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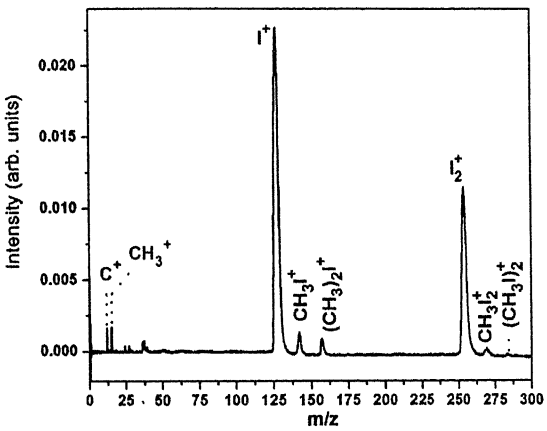

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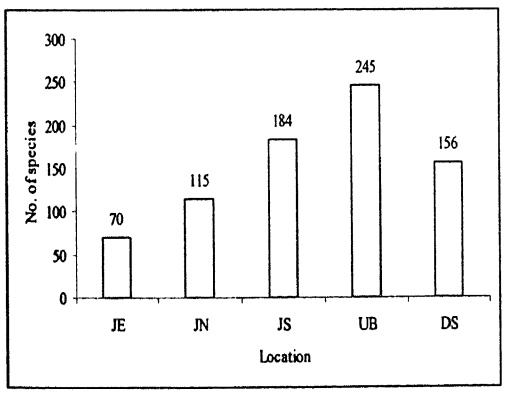

Entity relationship model to illustrate molecular evolution

S. Krupanidhi, S. Sai Madhukar and N.S. Umanath

The entity relationship (ER) modeling grammar is a tool for the illustration and effective communication of conceptual designs of commercial/business systems. Hitherto, ER models are being used in the computer science and information systems disciplines.

Short Research Communications

Aconitine alkaloids from tubers of *Aconitum heterophyllum* and *A. balfourii*: Critically endangered medicinal herbs of Indian Central Himalaya

H. Pandey, S.K. Nandi, A. Kumar, R.K. Agnihotri and L.M.S. Palni, F.N.A.Sc.

Population (Place)	Altitude (m)	*A. heterophyllum*	*A. balfourii*	
		Aconitine	Pseudoaconitine	Aconitine
Garhwal Himalaya				
Bharnala	3000	0.72	0.24	0.28
Doditul	3200	0.48	0.46	0.33
Goi	3200	NA	0.24	0.81
Dayara (Syuri Bugyal)	3280	0.30	0.33	0.68
Hemkund	3300	NA	0.45	0.79
Tungnuth	3600	0.14	NA	NA
Kedarnuth	3600	0.69	0.39	0.13
Kumaun Himalaya				
Khatiu	3250	0.67	NA	NA
Phurkia	3260	0.75	NA	NA
Kafni	3400	0.13	0.06	0.17
Phurkia Bugyul	3430	NA	0.62	0.83

NA= Not available at the time of collection; all values are average of 3 HPLC injections.

Quantification of major diterpenoid alkaloids, namely aconitine and pseudoaconitine was carried out in tubers of two Himalayan aconites collected from higher altitudes of Kumaun and *Garhwal* region, following column, thin layer and high performance liquid chromatography. The levels varied markedly amongst populations and the active principle content could not be correlated with the altitudinal difference

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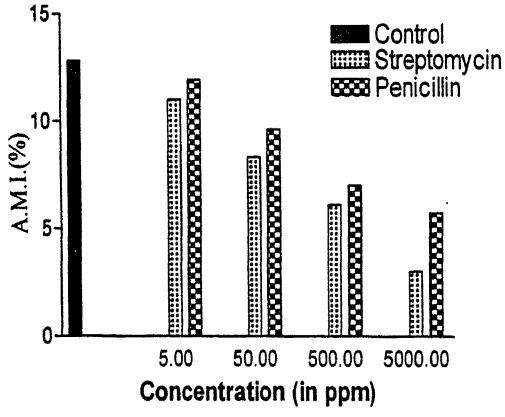
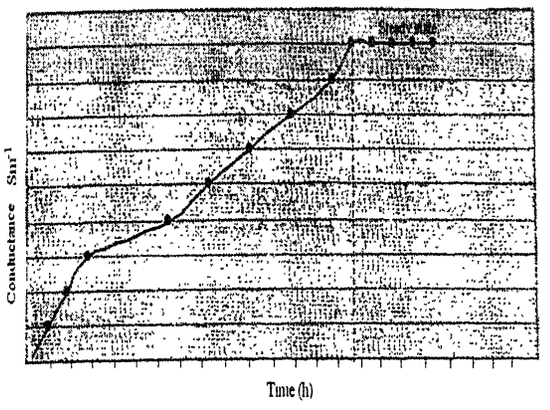


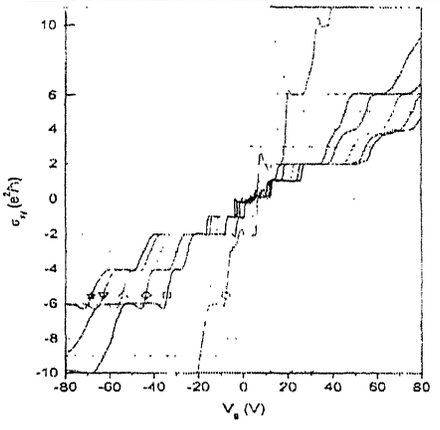
Clastogenic assessment of two antibiotics on safflower

G.Kumar and Preeti Srivastava

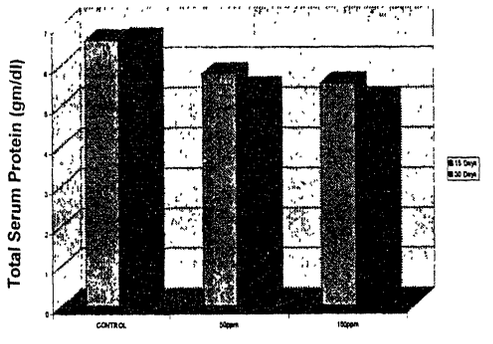
The graphs clearly show the comparative impact of the two antibiotics streptomycin and penicillin on A.M.I and chromosomal abnormalities. Though both the antibiotics displayed a decreasing trend of A.M.I and increasing trend of chromosomal abnormalities alongwith increasing concentrations but streptomycin proved to be more effective.



The new theory and diffusion technology developed earlier has been employed for seed identification, determining critical time for germination, water diffusivity and enhancing germination for Rare-1 and Melkassa-1 maize genotypes. The results showed 100% germination whereas diffusivity and water absorption for Rare-1 was 1.07 and 1.2 times higher than melkassa-1.



The substitution of the g value in the harmonic oscillator type of levels produces both the fractional as well as serial quasiparticle charges which are in agreement with the experimental values.



In the present study effect of ammonia gas inhalation on the serum protein level of albino rat was studied.

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Cover Page Photograph : Endemic plant *Hypericum gaitii* Haines p. 77.

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EDITORS' PAGE

Most, if not all, of the R & D Government Laboratories created under the banner of CSIR, ICAR, ICMR, DST, Department of Biotechnology / Electronics etc with massive inputs over the last 2-3 decades appear to be failing in fully delivering the much hyped quality basic and industrial research output. Poorly trained, un-motivated and non-committed young research personnels from the Universities/Centres of Higher Learning joining the R & D laboratories have always come out to be the essence of all the "analysis" done in the past to pin-point the reasons behind the lack-luster performance of various laboratories. Whom to blame? There is no use in playing the "blame - game". Our Academy, at different forums including this journal's Editorials / Articles / Seminars etc, have been emphasizing the dangers and negative effects of neglecting Universities as far as the fulfillment of long term R & D goals of the nation are concerned. Unfortunately, before something could be done in this regards, we have come at the cross-roads where lobbying in favour of strengthening University Education infra-structure is superfluous and no more required. ***Problem has already come face-to-face glaringly.*** *The emergent situation regarding the need of well trained University graduates / researchers is self-evident. Thus, once again, the education rightly takes the center stage of national development.*

Good quality teachers, committed students and infra-structure constitute the integrated education system whose improvement requires fast" intervention". The "hatching period" for producing good quality teachers / students is long and hard as compared to the infra-structural development. Therefore, a sustained effort with a well thought out approach is necessary without "panicking".

Let us evaluate the scenario regarding teachers in the University / Professional institutions. Some of the problem areas are: (i) Vacant teaching positions (ii) number of posts being less than desired optimum (iii) recruiting ad-hoc teachers on contract (iv) poor emoluments and working environment (v) Non-availability of qualified and inspiring teachers (vi) teachers with outdated knowledge and dis-interest in research (vii) Lack of " system-check" on the performance and accountability of the teachers. Some of the above are interlinked and many are even self-inflicted. Dealing with all these complex issues in one go may make a big document which is not the intention of this Editorial. Nonetheless, we do wish to emphasize that the quality of teachers should not be compromised in a state of "panic to recruit them fast" A single dead fish can make the whole pond dirty. Same is true for the presence of quality compromised teachers in the Universities/Centres of Higher Education.

Politicisation, partisan attitudes of authorities, unimaginative selection /qualification criteria, in-breeding and active "teacher pressure groups" are major culprits leading to compromises in the selection of teachers. These cannot be stopped completely but their effect can be minimized by fixing minimum qualification requirement" judiciously which have seen dilutions even in many reputed institutions. While good academic record, character and communication skills are "musts", the academic degrees of the teacher being recruited at the lowest level should be higher than the highest degree offered by the respective

department/institution. We are not going into the details because the requirements of Basic Science/ Technology/Medicine/other professional course streams are widely different. Some suggestions are given below which may act as the starting point for discussions: (a) For science stream institutions granting upto Ph.D. degree-M.Sc., Ph.D. with two years Post-Doctoral experience; good research publications. (b) For science stream institutions granting upto M.Sc. degree-M.Sc., Ph.D. (c) For medical stream-M.D./DNB with work experience and M.Ch. for departments with super speciality (d) Engineering institutions offering degree upto B.Tech. only-M.Sc., Ph.D. or M.Tech (e) For Engineering institutions offering M. Tech/ Ph.D-M.Tech/M.Sc. and Ph.D. with publications or work experience. The qualifications ought to be kept same for "Reserved Category" as well with minor reduction in the level of early academic records. It may be argued that by and large, presently existing qualification criteria are almost similar to those presented above. This is not true! The compromise in the form of having teachers with lower qualifications (like with bare B.Tech. degree only) in Engineering education is rampant which is eroding the credibility of its degrees fast, particularly with the mushrooming of Private Engineering colleges. We need not" panic" if some of the posts remain vacant because of the lack of good applications and/or even if some institutions come to the brink of closure. We realise that it is easier said than done. But corrective open surgeries are always painful. We have to gear up ourselves for better future!!

Other issues related to teacher recruitment and teacher's problem at the University level may be raised by us in subsequent Editorials. In the meantime, we request our readers and learned Fellows to sent to us their opinions and suggestions where full or abridged version may be presented, through these journals to the scientific community at large for their consideration and discussion.

Girjesh Govil
Jai Pal Mittal
Suresh Chandra

The views expressed here are solely those of one of the Editors and do not necessarily reflect those of the Academy or the Institute where he works.

Genomic sciences and medical biotechnology

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Abstract

The advent of genomic sciences has provided enormous information on the gene content and its expression in a wide variety of organisms, including the human. This has paved the way for understanding the differences between normal and abnormal cells at the molecular level. This enables molecular characterization of the abnormality due to a wide variety of causes ranging from infection to life style disorders. This new knowledge is being exploited to develop molecular diagnostics, new drugs and vaccines, biopharmaceuticals, gene therapy and stem cell therapy. The future of Medicine would be influenced by Genomic Sciences in a major way.

Keywords : genomic sciences, molecular diagnostics, gene therapy

The advent of genomic sciences has ushered in a top down systems biology approach to understand and treat diseases. The conventional knowledge or hypothesis driven approach based on specific genes and their products is still relevant and essential, but the systems biology approach enables identification of incriminating factors on a genome scale and creates a wider basket. However, the knowledge - driven bottom up approach is essential to sift the voluminous data and to provide experimental validation (Fig. 1).

Genomic sciences in a broad sense would include Genomics, Functional Genomics and Proteomics (Table 1). Genomics refers to the DNA/RNA sequence of the genomes of different organisms, assembly of the sequences to provide a gene map including exons, introns, repetitive sequences, Single Nucleotide Polymorphisms etc. Functional Genomics is based on the annotation of the genes in the genome. This is a major challenge, since not more than 40% of even *E. coli* genome is interpreted in terms of functionally known products, the rest being annotated as Open Reading Frames or

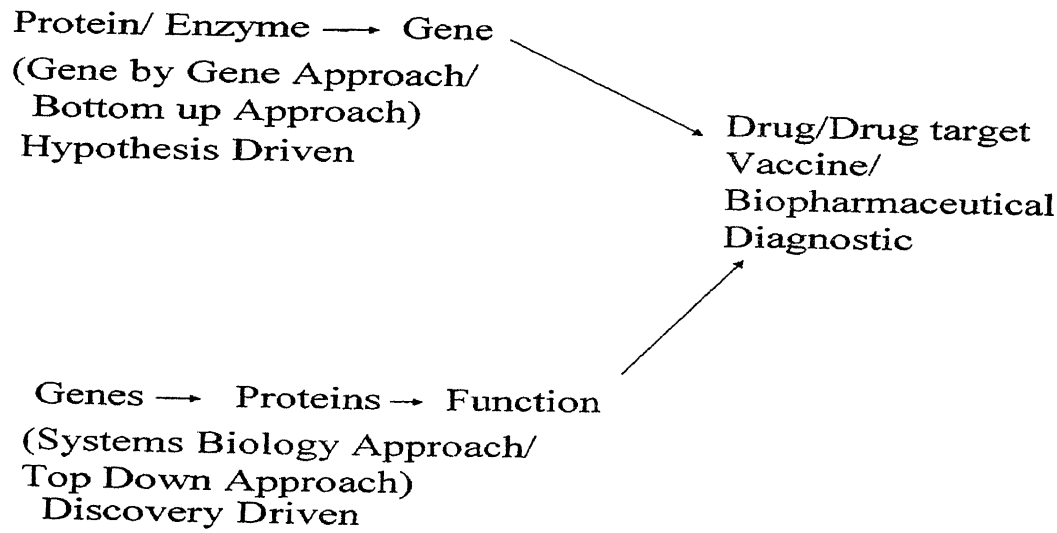


Fig. 1–Genome revolution.

Hypothetical Proteins. Functional genomics would include gene expression and its analysis using DNA microarray and transcription studies. Proteomics includes protein identification, quantification, localization, structure, protein-protein interaction, post-translational modification etc. Obviously a study of proteomics is much more complex than functional genomics, and the ultimate functional correlation of the proteins with metabolism defines the physiology or pathology.

Table 1– Genomic Sciences.

- Genomics
- Functional Genomics
- Proteomics

The complex instrumentation required for genomic sciences would include DNA sequencing machines, DNA microarray, 2D-Gel-electrophoresis of proteins, liquid chromatography, mass spectrometry, X-ray and NMR machines, protein purification and sequencing instruments, yeast 2-hybrid and other systems to study protein-protein interactions and a wide variety of bioinformatics tools. In order to obtain meaningful and manageable data, genomic sciences are now used at the level of sub-systems for genes and proteins, as for example those involved in signaling pathways or with respect to a specific cancer or a specific tissue or a specific family such as protein kinases.

The availability of sequence infonnation on the human and 200 other genomes has opened up new strategies for diagnosis and therapeutic interventions. These are briefly discussed below.

Molecular Diagnosis of Diseases:

Polymerase Chain Reaction (PCR) has become a powerful tool to identify the infectious agent in blood or other body fluids as well as to detect specific mutations, single nucleotide polymorphisms (SNPs) and other genome changes seen in genetic disorders, cancers etc. Similarly, DNA micro arrays (gene chips) can be made use of to identify cancer subtypes and other tife style disorders. Many attempts are underway to correlate SNPs to disease susceptibility. These highly sensitive methods have potential applications for prenatal diagnosis. While, these methods are still evolving, methods based on ELISA and related techniques using monoclonal antibodies to detect and quantify specific antigens are widely practiced.

New Drug Development Paradigm :

Leads from the genomic sciences have helped to identify specific drug targets as for example in metabolic and signaling pathways leading to drug development through rational drug design. High throughput screening techniques with an array of molecules against specific genes and gene products have been developed to identify candidate drug molecules. Apart from identification of specific molecular targets, recombinant DNA methods are also used to produce large quantities of the target proteins using a variety of expression systems (Table 2).

Table 2– New Drug Development Paradigm

1. Identification of Targets —Biochemical Pathways, Signaling Pathways, Leads from Genome Project.
2. Combinatorial Chemistry.
3. Molecular Modeling
4. High thruput screening.
5. Trials with candidate molecules.
6. Pharmacogenomics
7. Drug delivery using nanoparticles.

The area of phannacogenomics has become very important in the process of drug discovery and development. It is not well known that even the most successful drug in the market cures only 40% of the population optimally. This is because of variations in the genetic potential of individuals to absorb, metabolise (eg. cytochrome P-450s and Phase II drug metabolizing enzymes) and excrete drugs, in addition to molecular changes in the receptor target. The subject of pharmacogenomics deals with genes involved in the processes mentio-

ned, besides also throwing light on such changes during drug-drug interaction. An ideal situation would be to move away from fixed dose formulations to designer drugs, perhaps an utopian dream at this stage. Pharmacogenomics would atleast help to identify subpopulations that may not respond to a drug optimally and such knowledge would be useful to the drug company as well as the patient.

Molecular Medicine

This would include therapy or protection based on protein pharmaceuticals including vaccines, nucleic acid based therapies and cell-based therapies (Table 3). Insulin, growth hormone, interferons, growth factors etc are classical examples of protein pharmaceuticals. Cell culture based and recombinant protein based vaccines represent the evolution of vaccine technology from the classical whole-organism based inactivated or attenuated vaccines. The advent of DNA vaccines has revolutionalised the field of vaccinology. A prime boost approach involving a DNA vaccine primary followed by a viral vector expressing the gene of interest as a booster is now widely tested for scores of diseases. Nucleic acid based therapies hold tremendous promise for the future. These include gene therapy for genetic disorders, a few examples of which are listed in Table 4. Around 5000 genetic disorders are known to mankind and as more and more disorders are identified at the gene level, such genes would become candidates for gene therapy. Although germ-line therapy is not ethically feasible in the human, somatic cell gene therapy is a distinct possibility, although issues of gene targeting to specific cell types, efficiency of gene transfer and regulated expression are still major challenges to be . overcome. Viral-mediated gene transfers are more efficient than physical methods of gene transfer. However, the former approach carries the risk of vector-mediated side effects. Although, successful results have been obtained with Severe Combined Immunodeficiency Syndrome (SCID) in the human, the challenges already mentioned need to be overcome before gene therapy can become a clinical practice for different genetic disorders.

Table 3–Molecular Medicine.

PROTEINS	Cell Therapy
Hormones	Stem Cells
Enzymes	
Growth Factors	
Immunomodulators	
Monoclonal Antibodies	
Vaccines	
NUCLEIC ACIDS	
Genes for Therapy	
Antisense Oligonucleotides	
Ribozymes	
DNA Vaccines	

Table 4–Single Gene defects — current targets for gene therapy (examples)

Disease	Defective Gene
Cystic Fibrosis	CFTR
Pituitary Dwarfism	hGH
Emphysema Familian	α–1 antitrypsin
Hypercholesterolaemia	LDL Receptor
β–Thalassemia	β–Globin
Sickle cell anaemia	β–Globin
Haemophilia A	Factor VIII
Haemophilia B	Factor IX
Gaucher's Disease	Glucocerebrosidase
Phenylketonuria	Phenylalanine hydroxylase
SCID	ADA, Purine nucleotide–Phosphorylase
DMD	Dystrophin

Molecular Medicine in the case of cancer therapy has involved approaches based on the use of antisense oligonucleotides to prevent the expression of oncogenes such as c-ras, c-myc, c-myb etc in

Table 5– MAbs used in cancer therapy.

MAb	Trade Name	Used to Trast :	Approved in :
Rituximab	Rituxan	Non-Hodgkin lymphoma	1997
Trastuzumab	Herceptin	Breast Cancer	1998
Gemtuzumab ozogamicin*	Mylotarg	Acute myelogenous leukemia (AML)	2000
Alemtuzumab	Campath	Chronic lymphocytic leukemia (CLL)	2001
Ibritumomab tiusetan*	Zevalin	Non-Hodgkin lymphoma	2002
Tositumomab*	Bexxar	Non-Hodgkin lymphoma	2003
Getuximab	Erbitux	Colorectal cancer	2003
Bevacizumab	Avastin	Colorectal cancer	2004

*conjugated monoclonal antibodies

experimental tumors. Strategies to enhance P53 expression, since P53 is depleted in 50% of the cancers as well as to specifically lyse P53 negative cells by a modified adenovirus (ONYX - 015) have given encouraging results. The most successful therapeutic option has been the use of MABs against specific cancers listed in Table 5. Autologous cancer vaccines, where the patient's own cancer cells are genetically engineered with cytokines and growth factors to enhance their immunogenicity, have also given encouraging results. Similar strategies are also being investigated against cardiovascular disorders. Cell therapy needs to be considered as a part of Molecular Medicine. Diseases involving wholesale degeneration of cells may not just respond to the replacement of a few genes, but would necessitate the replacement of cells themselves. It is here that stem cell therapy holds tremendous promise to generate different types such as neuronal cells, cardiomyocytes, hapatocytes etc. Embryonic stem cells are considered superior to adult or cord blood stem cells, since they have the potential to generate wider range of cell lineages. However, the latter have direct applications in hematopoietic cell regeneration. The use of embryonic stem cells needs further understanding of their genetic potential under different conditions of maintenance to retain 'stemness' as well as to generate specific cell lineages. Targeting of stem cells to specific tissue locations is another challenge (Table 6).

Table 6–Stem Cells.

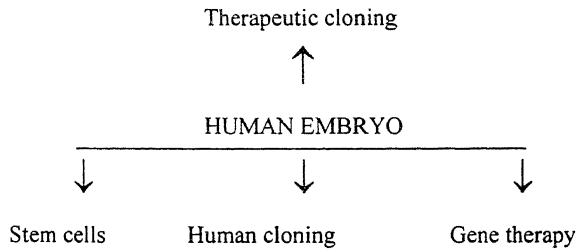
•	Factors influencing cell plasticity
•	Injury
•	Cell type
•	Timing of engraftment
•	Chimerism with radiation
•	Route of administration
•	Number of cells administered
•	Functional state of cells
•	Stem cell mobilization after engraftment
•	SCIENCE 308, 1121, 2005

Ethical Issues:

The future of Medicine holds tremendous promise with the application of new knowledge from genomic sciences. At the same time, there are ethical issues that need to be considered. Prenatal molecular diagnosis carries its own load of ethical concerns. Often, diagnosis without the availability of a cure

can only bring in more stress to those concerned. Medical termination of pregnancy under such situations is not universally accepted. Germline gene therapy has still not evolved to confidently ensure the well being of the embryo in its natural state. The consequence of introducing a foreign gene to a chromosomal location that is not its natural locus can be serious. This issue has come up seriously with even somatic cell gene therapy. The remarkable success with SCID–XI patients is laced with the sad development of leukemia in 3 out of the 17 children. The long term effects of gene therapy or even stem cell therapy need to be understood, if they have to be used in children and young adults. The human embryo has now occupied the centre stage in all ethical discussions, since it is the target in germ line gene therapy or prenatal diagnosis or preparation of embryonic stem cells, besides its central role in reproductive or therapeutic cloning (Table 7). The source of the human egg cell even for therapeutic cloning is an issue. While, the debate on the ethics of using or manipulating the human embryo goes on, there is no doubt that the future of Medicine will be largely influenced by the genomic sciences.

Table 7–Ethics/morality.



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Photochemistry of Clusters

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Abstract

This article gives a brief introduction to the field of atomic and molecular clusters. In brief, classification of clusters and different methods used for their generation and characterization have been described. Interesting new photochemical aspects of cluster as compared to its monomer have been discussed. We conclude with a potential application of cluster in the form of Coulomb explosion, a method which can provide a source of highly energetic multiply charged ions, electrons, neutrons and higher order harmonics.

Keywords : photochemistry, atomic and molecular, crystals, coulomb explosion

Introduction

In the last few decades, the field of cluster science has become a frontline research area of interdisciplinary interest, due to advancement in theoretical and experimental techniques. These theoretical and experimental methods have helped in understanding the geometric and electronic structure of clusters, as well as in engineering the properties of these novel materials for specific technological applications by changing size and composition of clusters[1-3]. Because, unlike in liquid or solid, in clusters nearly all of the atoms/molecules are on or near the surface. Due to this surface effect, reactivity of a cluster can be significantly altered with the loss or addition of a single atom/molecule. This results in size dependent changes in electrical, magnetic, optical, chemical and catalytic properties. For small cluster, these variation are very strong and not a linear function of size. For larger clusters, the dependence on size gets gradually weaker and there is smooth development of properties towards those of infinite solid. The size dependence of chemical and physical properties of clusters has provided a convenient handle to scientists for tuning the cluster properties to desired value, by changing the cluster size[4]. Also, due to their ability to provide bulk-like density in gas phase, clusters have been employed as nano-laboratories for basic understanding of different physical, chemical and biological problems - like energy dissipation, influence of solvation, interaction between biomolecules etc.[5-7]. Besides being of academic interest, study of clusters has important implications in the field of homogeneous and heterogeneous catalysis and solid state physics.[8,9]. As reactions on clusters have analogies with reaction on surfaces and are important for understanding catalysis, etching and adsorption.

Similarly in the field of chemical physics, availability of laser armory and generation of clusters in collision free environment isolated from background by supersonic expansion, has ignited studies on clusters for fundamental understanding of intermolecular interactions and structures, energy dynamics, effect of solvation and photochemistry of clusters. Besides these, photochemical/spectroscopic studies are important for unraveling the weak attractive forces that bind different molecules within the cluster.

From the glimpses presented above it is clear that the topic of clusters is very broad, with varied scientific interest. However, in this review our main focus would be on photochemistry of vander Waals clusters and how their behaviour differs from that of its constituent monomer unit. But before we focus on this topic we would first like to give a broad basic introduction to the subject of clusters.

Clusters and their classification

Clusters are defined as aggregates of atoms/ molecules, commonly intermediate in size between

individual atoms/molecules and aggregates large enough to be called bulk matter[10], which are held by interactions ranging from weak vander Waals forces to strong ionic bonds. These clusters are formed by bringing atoms/molecules together in such a way that the forces holding them do not saturate, so that the number of constituents in the clusters can be altered deliberately, without significantly changing the local structure and properties of the clusters. This characteristic, which may be regarded as a primary property of clusters is referred to as stackability. As mentioned previously, the forces which hold these atoms/molecules within the cluster are of different types ranging from weak vander Waals interactions to strong Coulombic interactions (see Table 1). The structure, stability, reactivity and properties of clusters strongly depended on the type of interactions, which hold the constituent species within the cluster.[11]. Depending on the nature of constituting atoms and binding characteristics, clusters can be broadly classified into following categories :

(a) *Molecular cluster:* These clusters are formed by the agglomeration of molecules without disturbing their composition. The type of bonding exhibited by molecular clusters includes vander Waal bonding, dipole-dipole interactions, higher order multipolar interactions and hydrogen bonding. In these clusters the most prominent interaction is of vander Waal type as each molecular species is highly stable and weak interactions are expected between two molecules. Examples of molecular cluster are $(N_2)_n$, $(C_6H_6)_n$, $(CH_3I)_n$, $(HF)_n$, $(H_2O)_n$.

(b) *Rare gas clusters:* At low temperatures, it is possible to form clusters of rare gases like Ar, Ne, and Kr. These clusters are bound by weak vander Waals type forces. The interatomic attractive force increases with increasing atomic mass. The strength of binding energy is of the order of ~ 0.3 eV/atom or less (1 eV= 23.06 kcal mol^{-1}).

(c) *Ionic clusters:* Ionic clusters are also called hetero-atomic clusters as they are formed from the atoms with large difference in the electronegativity. Since the anions and cations have fixed charges, the overall charge of the cluster depends on its stoichiometry. The bonding is strong with a binding energy ≈ 2-4 eV/atom. In this class, we have the clusters like $[Na_xCl_y]^{(x-y)+}$, $[Mg_xO_y]^{2(x-y)+}$ etc.

(d) *Semiconductor clusters:* These clusters are aggregate of elements which are semiconductor in nature like Si, Ge and other compounds like GaAs, CdTe. In this type of clusters, the bonding is covalent in nature i.e. bonds are highly directional and strong. The binding energy in semiconductor cluster is typically of the order of 1 to 4 eV/atom.

(e) *Metal clusters:* These clusters consist of elements, which exhibit metallic properties in bulk phase i.e. Na, Al, Cu, Ag, Au etc. The interatomic forces in metallic clusters are a combination of many forces and the nature of bonding depends strongly on the size of cluster. The strength of the binding in metallic clusters ranges from 0.5 to 3 eV/atom. The properties and bonding in metal clusters vary significantly for metals of different blocks of the periodic table. For clusters of transition metal clusters we would like to emphasize regarding their magnetic properties as a function of size. Although most transition-metal atoms have unpaired d-electrons and are magnetic, very few transition-metals are magnetic in bulk. In studies carried out on clusters of iron, cobalt and nickel clusters using Stern-Gerlach molecular beam deflection method clusters of iron, cobalt and nickel were found to be superparamagnetic with strong size-dependent magnetic moments. In these studies, smaller clusters (fewer than ~30 atoms) were found to have atom-like magnetic moment, while as the size of cluster is increased to ~700 atoms the magnetic moment was found to approach bulk limit.[12]. Also, magnetism has also been detected in clusters of some elements which are nonmagnetic in bulk phase.[13] e.g. - though ferromagnetic behavior has not been reported for pure 4d transition metals, but clusters of rhodium, Rh_n (n=12–32) display giant magnetic moments. Similarly, although bulk Pd metal is diamagnetic in nature, its clusters of finite size are found to show significant magnetic moment.

Table 1– Classification of clusters

Type	Example	Nature of interaction	Binding energies
Ionic	$(NaCl)_n$	Ionic	~2-4 eV
Covalent	C_{60}	Covalent	~1-4 eV
Metal	Al_n	Metallic	~0.5-3 eV
vander Waals	Rare gas clusters, $(CO_2)_n$, $(H_2O)_n$, $(C_6H_6)_n$, $(CH_3I)_n$, $(HF)_n$,	Polarization, Hydrogen bonding	≤ 0.3 eV

In this review, we will be dealing mostly with molecular clusters. In some of these, such as $(CH_3I)_n$, in addition to vander Waals interactions hydrogen bond interactions also contribute in the formation of clusters. These hydrogen bonded clusters are held more tightly than vander Waals clusters but less tightly than the covalent and ionic clusters.

How clusters differ from molecules?

In molecules also different atoms are held together by ionic and covalent interactions. Hence, it is necessary to ask, how clusters differ from the molecules? Molecules are stable and have a definite stoichiometry and structure, since the atoms in a molecule are bound by rigid and directional chemical bonds. On the other hand, clusters are meta-stable and do not have definite composition, since their composition strongly depends on production condition and can be varied by changing experimental parameters. In addition due to their open structure, clusters can bind more and more atoms/molecules and grow larger without any size limitation. Thus, in contrast to the well-defined rigid geometrical structure of molecules, a cluster often has a modular structure.[14] Also as compared to molecules, clusters have a large number of isomeric structures, degrees of freedom and closely spaced energy levels. Further, in contrast to molecules which generally always have a unique structure, the structure and properties of the cluster are dependent on its size (no. of atoms/molecules) and with increase in size the number of stable structures increases.

Having distinguished the clusters from molecules, it is pertinent to compare clusters with nanoparticles. Though these two novel states of matter are interrelated, there is a very thin line that separates the two, based on their size and properties as compared to bulk. Clusters in general can be categorized as small, medium and large (Table 2). Small clusters exhibit properties, which depend significantly on the size and shape of the cluster. In this size regime there are islands of magic clusters associated with closed shell structure, for these clusters the variation in properties cannot be correlated directly with the number of constituent units (atoms or molecules). While for medium sized clusters, though there is appreciable change in properties of cluster as a function of clusters size, but the variation is smooth and can be interpolated (traced) with the size of clusters. On the other hand - in the regime of large clusters, the observable properties slowly starts converging towards their corresponding bulk material (Fig. 1). These clusters have dimensions in the nanometric range, which qualifies them as nanoparticles as by definition nanoparticles are those materials which are of nanoscale in three dimension and which also exhibit new or enhanced size-dependent properties compared to the larger particles of the same materials. In these nanoparticles, the structure of their nucleus is similar to that of their bulk, but on the surface they exhibit different structure.

Table 2–

Clusters	Number of atoms/molecules (N)*	Diameter (d)*
Small	$2 \leq N \leq 30$	$d \leq 2$ nm
Medium	$30 \leq N \leq 500$	$2 \text{ nm} \leq d \leq 5$ nm
Large	$500 \leq N \leq 10^{6-7}$	$5 \text{ nm} \leq d \leq 100$ nm

*The size regions listed in table 2 are not unique, because no clear demarcation exists.

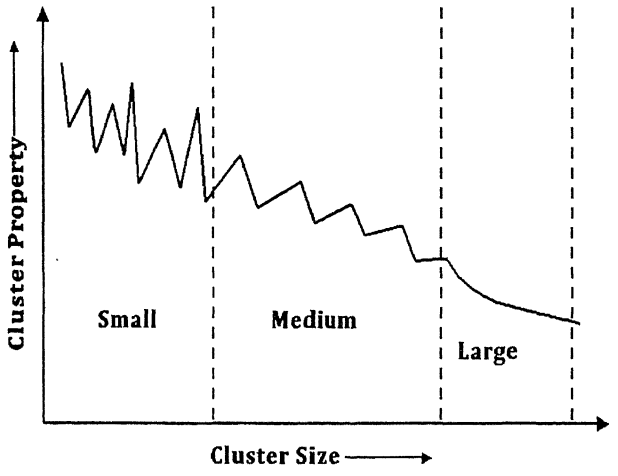


Fig. 1– Representative graph depicting variation in properties of cluster as a function of size

Current interest in cluster studies

As discussed earlier, clusters have been of interest to the scientific community due to their potential to offer ways to make altogether new kinds of material, as their structural and electronic properties bear resemblance neither to atoms/ molecules they are composed of, nor to the corresponding bulk matter they eventually get transformed to. In addition, atomic and molecular clusters offer a unique medium to understand, how the physical and chemical properties of matter emerge, as atoms and molecules associate together one by one. Although the properties of solid and liquids may be extrapolated to those of small fragments by scaling down, but this simple estimate is invalid in the cluster size regime. In particular, the properties of clusters change irregularly in a nonscalable manner with the number of constituent atoms or molecules in the cluster regime (Fig. 1). This size specificity is the most distinct feature of cluster, which is manifested most clearly in free clusters, isolated in the gas phase. The specific size of a cluster may be referred to as a magic number, when it is distinguishable from those of neighbouring sizes in properties such as abundance, reactivity, ionization potential, electron affinity, bond dissociation energy, etc. The primary factor determining properties of a cluster is its electronic and geometric structure, which are investigated by various experiments including laser spectroscopy, atomic collisions and surface impact, as well as by taking essential inputs from theoretical methods also.[5]

From technological point of view, clusters often exhibit unique physicochemical properties, which are not present in atoms, molecules or macroscopic bulk materials. These unique physicochemical properties of a cluster originate mainly from the fact that a cluster is composed of a finite number of atoms/molecules, most of which are located on its surface i.e. large surface to volume ratio. Moreover, with increase in size these small volumes rapidly approaches bulk behavior.[6] Thus, they can provide bulk-like conditions and at the same time, because of their finite size, a possibility to measure observables, inaccessible in the bulk (e.g., fragments of photolysis of a molecule in cluster can escape from the cluster, while they are usually trapped in the bulk). Therefore, cluster research often investigates physical and chemical properties as a function of cluster size ultimately resulting in understanding the transition from the molecule to the bulk.

It is also worth mentioning that the finite size of cluster allows theoretical treatment of these systems, which is not feasible for the bulk. These unique cluster properties have led to their use as nano-laboratories for investigations of various physical and chemical processes. Clusters provide a very efficient heat bath having low characteristic temperatures (~1 to 10's of Kelvin).[7, 8] For example, large He clusters are used for spectroscopy of embedded molecules and for investigation of chemical reactions in cold environments (~few Kelvin), while photolysis of molecules in rare-gas clusters, provides necessary input for understanding their behaviour in bulk for similar process.

Focus of this review

The topic of clusters is vast and diversified with different scientific and technological significance. In this review we will restrict our selves to molecular clusters. Because the linkage of these cluster systems and their processes to photochemistry that occurs in liquid and solid phase is principally strong. The term molecular clusters denotes aggregates of molecules ranging from dimers to conglomerates of thousands constituents, bound together non-chemically, e.g., by weak vander Waals interactions

or by hydrogen bonds, etc.[9]. These interaction avoid rigid bond formation and breaking, which would otherwise limit the stackability of molecular clusters. These molecular clusters are special in the sense that they involve two different types of binding: the weak intermolecular interactions, and typically an order of magnitude stronger chemical bonds of their constituent molecules. For example vander Waals clusters have binding energy per bond smaller than 100 meV, while hydrogen bonded clusters have binding energy per bond smaller than 300 meV. Though these intermolecular interactions within the cluster may be deemed as weak, but they have a profound impact on the properties of the clusters. Ono et al. have measured ionization energies of CS_2 molecule and its clusters as a function of clusters size. In their studies, as compared to ionization energy of 10.068±0.002 eV for the lower spin orbit state of CS_2, the ionization energy for clusters of CS_2 i.e. $(CS_2)_2$, $(CS_2)_3$, $(CS_2)_4$ and $(CS_2)_5$ were found to be 9.36±0.02, 9.22±0.02, 9.10±0.02 and 9.04±0.02 eV, respectively.[15] The point to note here is that though the vander Waals forces are of meV order, but the changes in physical and chemical interactions of clusters that these bring in could be much larger. In the following sections we will first give a brief general description of different methods for generation and characterization of clusters. Followed by an elaborate discussion on photochemistry of molecular clusters as compared to their monomer counterpart.

Different methods for preparation of clusters

Clusters are finite size microscopic objects. In order to produce them, one can either aggregate its smaller constituents (atoms, molecules) i.e. top up approach or break larger bulk systems i.e. bottom down approach. Clusters can also be made in solution in form of colloidal particles. However, to carry out studies on pure clusters (free from matrix) it is essential to prepare clusters in gaseous phase. Hence, we will concentrate mainly on the preparation of clusters in gas phase by different methods.

(a) Supersonic jet source

This dynamic method involving supersonic expansion of gases through a nozzle into vacuum provides a very convenient means of generating gas phase neutral clusters of atoms and molecules. The basic process involves adiabatic expansion of a gas or a gaseous mixture at high stagnation pressure (P_0) though a small orifice of diameter d into an evacuated volume having pressure in the range of 10^{-6}–10^{-5} Torr. The two body collisions during expansion converts random motion into directed flow, thus reducing translation, rotational and vibrational temperature of the molecule to few Kelvin.[16, 17] While, three body collisions coupled with extremely low temperatures and high number density of gaseous molecules leads to nucleation and condensation of clusters. These three body collisions are essential during the initial growth of the clusters. Since the process of cluster formation is exoergic in nature and the third body is required for removal of excess energy from the system in the form of kinetic energy. The process of cluster formation can be schematically represented by following equations-

$$M+M+X \rightarrow M_2+X \quad \text{(cluster nucleation)} \qquad (1)$$

$$\left.\begin{aligned} M_2 + M + X &\rightarrow M_3+X \\ &\vdots \\ M_{n-1} + M + X &\rightarrow M_n+X \end{aligned}\right\} \text{cluster growth} \qquad ..(2)$$

Here M represents the gaseous molecule, while X represents the carrier gas. Generally less condensable inert gases are used as carrier, because they efficiently remove the condensation/excess energy from the cluster system via collisions. The dimer acts as site for further growth and condensation. Need for third body participation is less for the later growth steps since the exoergicity can be soaked up by the cluster itself, particularly so when many vibrational degrees of freedom are available to act as the sink. Using this method, clusters can be produced as a molecular beam. The degree of cluster formation can be controlled by changing the nozzle diameter (d), stagnation pressure (P_0) and initial gas temperature (T_0). The rate of three body collisions (Z_3) and in turn the cluster formation is related to these parameters by the equation-

$$Z_3 \alpha P_0^2 d / T_0^2 \qquad (3)$$

This implies that cluster formation is facilitated by using large diameter nozzles and large reservoir pressure. The onset of clustering, and size of clusters produced can also be described by an empirical scaling parameter Γ^*, commonly referred as Hagena parameter[18, 19] and given by-

$$\Gamma^* = k\frac{(d/\tan\alpha)^{0.85}}{T_0^{2.29}}P_0 \qquad (4)$$

where d is the nozzle diameter (mm), α is the expansion half angle, P_0 is the backing pressure (mbar), T_0 is the pre-expansion temperature (Kelvin), and k is a constant related to bond formation. Clustering generally begins for $\Gamma^* > 100$-300, with the number of atoms per cluster N_c scaling as $N_c \propto \Gamma^{*2.0-2.5}$ [20, 21].

(b) The Pick-up source

The pick-up technique is an efficient method for preparation of embedded heterogeneous clusters, whereby a foreign atom/molecule/cluster is externally attached on the host cluster prepared by supersonic jet.[22] In a typical experimental setup, clusters generated upon supersonic expansion are crossed by a beam of foreign atom/molecule/ cluster just in front of the skimmer. Collision between the supersonically generated clusters and the external atoms/molecules results in the formation of heterogeneous clusters. This method is widely used for preparation of metal doped molecular clusters.

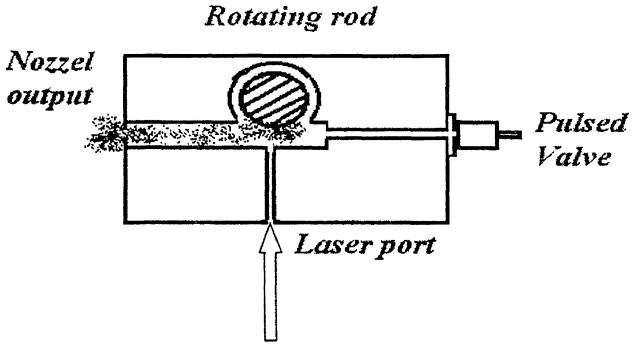


Fig. 2– Smalley type Cluster source designed at Chemistry division for generation of metal cluster by laser ablation.

(c) Laser ablation and vaporization cluster source

Both the methods mentioned above are well suited for preparation of cluster of volatile and low melting compounds. But the direct method of supersonic jet is not suitable for preparation of clusters of refractory materials, such as metals and ceramics which have low number density even at higher temperatures. For clusters of these refractory materials, use is made of laser vaporization technique coupled with supersonic expansion source. Advent of this laser vapourization technique enabled researchers to produce clusters of virtually any element in the periodic table and thus initiated extensive interest in the studies of clusters with varying composition, regardless of their volatility. Laser vaporization can generate a high number density of vapours of virtually any material in a short time interval and in a well-localized volume. By rapid quenching of the plasma using inert carrier gas, clusters can be produced. Hence, laser vaporization has become one of the most common techniques for generating cluster beams especially of refractory materials.[23] In this method, the refractory material whose clusters are to be prepared is used in the form of pellet or rod and ablated with a focused pulse of a laser beam. This results in generation of plasma at the target surface with temperature ~ 10^4 K. The resultant laser eroded material (atoms/small clusters) is supersonically cooled using monoatomic inert gases, resulting in formation of clusters of refractory materials (Fig. 2). A similar setup was used by Smalley and coworkers, for generation and characterization of carbon cluster, formed by supersonic cooling of plasma produced upon laser ablation of graphite. The resultant carbon clusters produced were detected using time of flight mass spectra. In their experiment they observed that under certain expansion conditions the mass peak corresponding to C_{60}^+ ion was the most dominating peak in the mass spectra, which later led to discovery of Buckminister fullerene having a truncated icosahedral structure made up of 12 pentagonal and 20 hexagonal rings.[24] Hence, such a laser vaporization cluster source is also referred as Smalley type source.

Having described different methods for generation of clusters, we would also like to mention that

production and characterization/analysis of clusters are interrelated and a single cluster setup houses the clusters generation source as well as the analyzer and the detector for characterization of the cluster properties. Fig. 3. illustrate a schematic diagram of the cluster setup designed and fabricated indigeneously at Chemistry Division, BARC, while Fig. 4. gives a pictorial view of the same. Here a pulsed valve mounted in the expansion chamber acts as a cluster source for generation of molecular clusters. In the present setup for formation of clusters, a carrier gas, typically an inert gas (helium or argon) at a stagnation pressure of 1-8 bar is bubbled through the liquid sample. The resultant gas sample is supersonically expanded through a pulsed nozzle, which results in the formation of clusters. The resultant supersonic jet is skimmed with the help of a skimmer (a truncated cone) and a cold beam of molecular clusters are introduced in the analyzer chamber. In the analyzer chamber these clusters can be characterized by different techniques (as discussed below). As our interest centers around understanding the photochemical behaviour of clusters upon interaction with laser pulses and to compare how it differs from its monomer counter part. In our case, we irradiate the clusters with photons and analyze the resulting electrons and ions produced. Accordingly, in the present setup cluster are subjected to focused laser pulses and the resultant cluster photofragment and ions produced upon multiphoton dissociation/ ionization of cluster are identified by time of flight mass spectrometer.[25-27]

Different methods for characterization of clusters

All the cluster sources produce a broad size distribution of clusters. But as properties of clusters

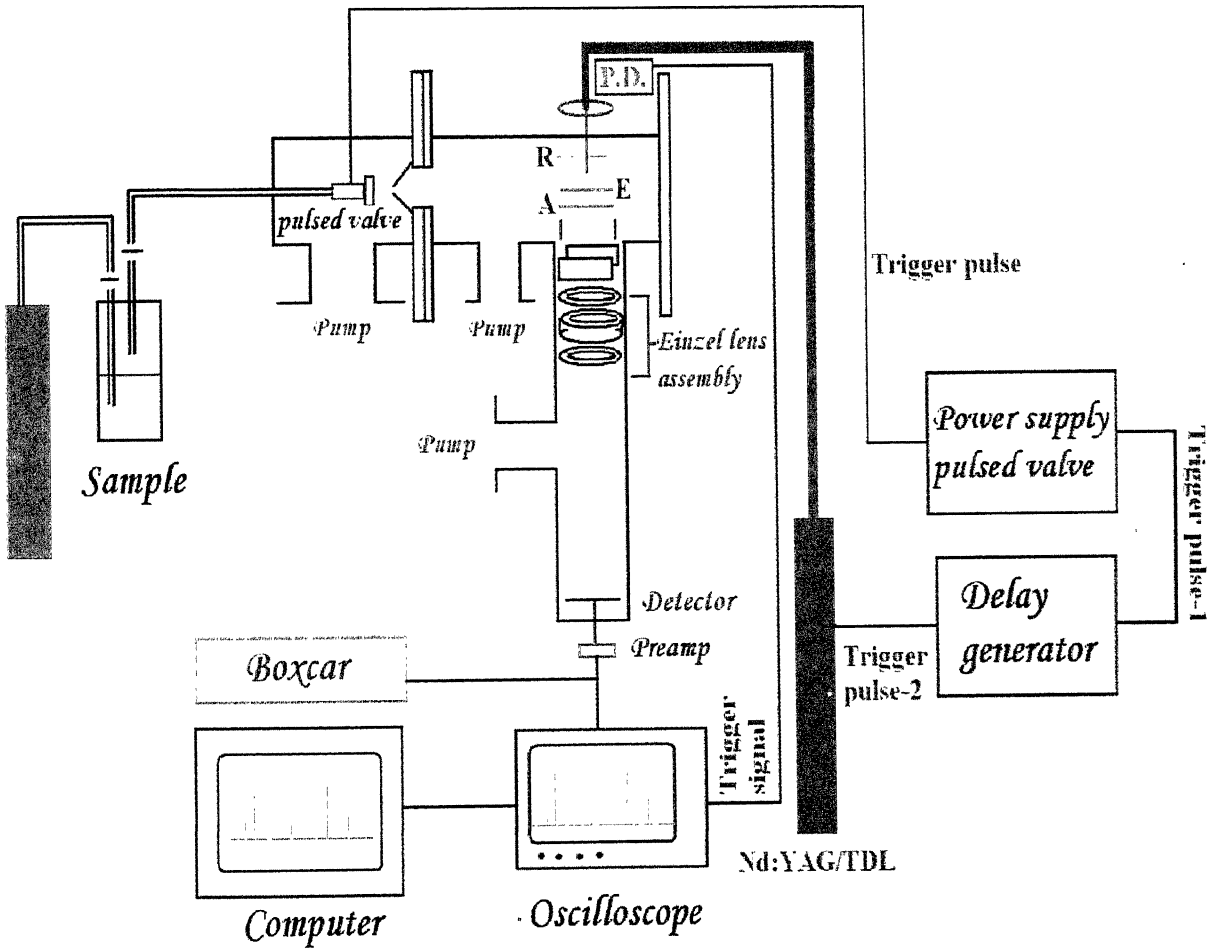


Fig. 3- Schematic layout of cluster setup designed and fabricated at Chemistry Division, Bhabha Atomic Research Centre-Trombay.

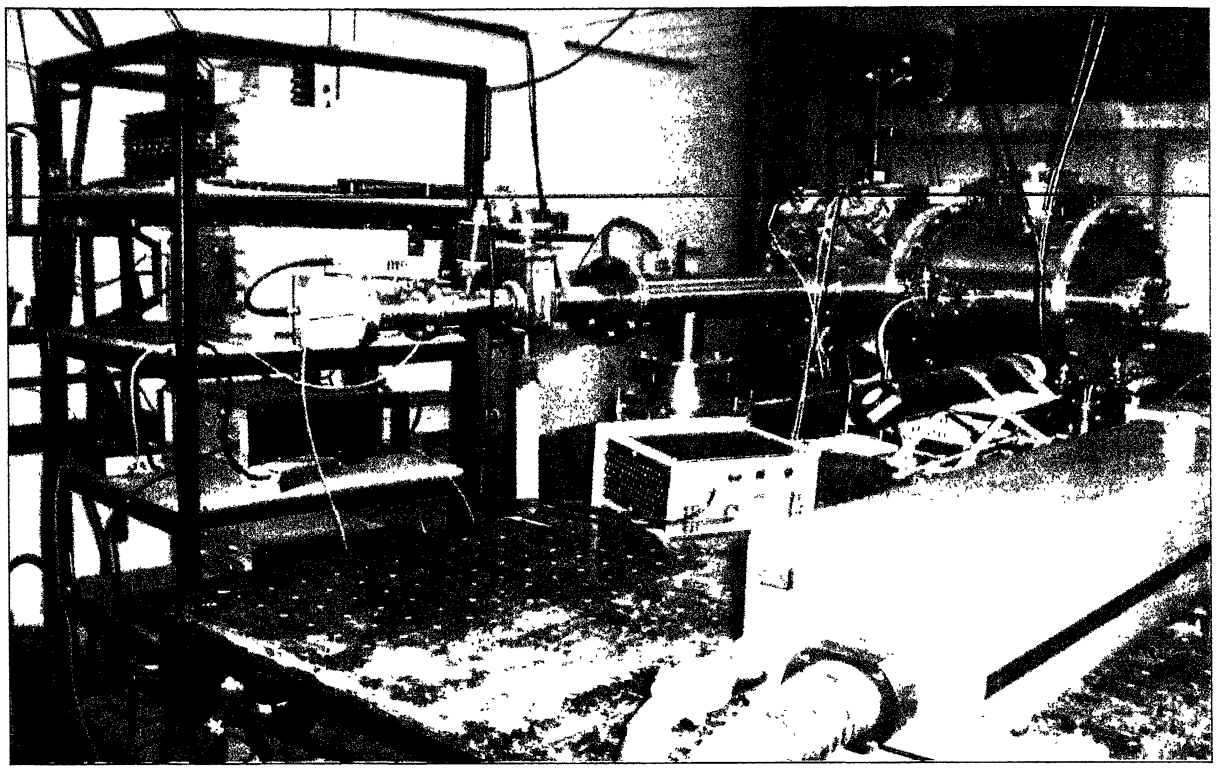


Fig. 4- Pictorial view of cluster setup designed at Chemistry Division, Bhabha Atomic Research Centre-Trombay.

depend on the size of the cluster i.e. the number of its constituents, it is essential to characterize the clusters for their size distribution. Since cluster properties are sensitive to size, methods should be devised to isolate clusters of definite size for specific applications and studies. Though mass spectrometer is widely used for isolation of size specific cluster ions, they are hardly used for measurement of mean cluster size and cluster size distribution of neutral clusters in a cluster beam. This is mainly because clusters undergo facile fragmentation and evaporation due to low binding energy of its constituents during the primary ion formation stage in the mass spectrometer. In addition, mass selective yields of ion detector in the mass spectrometer also limits the use of mass spectrometer for size characterization. Traditional technique of electron impact ionization (70 eV) followed by mass spectrometric detection of cluster ions leads to erroneous interpretation of cluster size distribution as the clusters undergo heavy fragmentation by energetic electrons. Similarly the method of laser ionization too suffers from the serious disadvantage of evaporation/ fragmentation and one can rarely detect the highest mass. Hence special techniques have been developed to characterize size of clusters in a supersonic beam.

For measurement and characterization of cluster size distribution, use is made of techniques such as Rayleigh scattering (Photon scattering) and electron diffraction. While atom scattering and mass spectrometric isolation of cluster ion (followed by reneutralization) is the most widely used technique for separation of cluster of specific size. Similarly, properties of small cluster have been studied using techniques such as IR and UV methods, as they are more close to molecules. While for studying large cluster properties, use is made of techniques related to solid state physics. These different techniques are briefly described below.

(a) Rayleigh scattering

It is difficult to measure the size of clusters produced in a free jet expansion directly, however its mean size can be estimated using Rayleigh scattering.[28] This method gives an average picture and conveys nothing about the distribution of cluster size. In order to estimate (obtain an approximate idea) the sizes of the cluster produced by the supersonic source, generally use is made of Rayleigh scattering measurements of clusters. Since the scattering signal scales as λ^{-4}, for carrying out Rayleigh scattering experiments, generally short wavelength laser pulses (in UV region) are preferred. These measurements are based on the fact that Rayleigh scattering cross-section scales as R^6, where R is the radius of the cluster. In these studies the Rayleigh scattering signal (S_{RS}) is expected to scale as P^3, where P is stagnation pressure[28]. In a typical experiment used to carry out Rayleigh scattering experiments, the UV laser light is crossed with the supersonic jet ~2-3 mm downstream from the nozzle.[28] In these experiments, it is necessary to avoid stray light from the incident laser since the scattered light is at the same laser wavelength. Hence, blackening of the inside chamber wall and use of Brewster windows for entry and exit of incident laser are very essential to avoid stray scattering.

(b) Electron diffraction method

Electron diffraction studies provide useful information regarding the mean cluster size. In addition to that, these studies give valuable information regarding the mean cluster geometry and cluster temperature.[29, 30] In a typical electron diffraction experiment, the supersonic cluster beam is interacted with a well-collimated electron beam with large kinetic energy (25 to 60 KeV). These electrons construct a diffraction pattern similar to Debye-Scherrer rings, on scattering from the atoms/molecules of the cluster. The shape of Debye-Scherrer rings in turn provides information regarding the mean size and other properties of the cluster. This method has been applied for determination of mean size of cluster in supersonic expansion of CO_2, Ar etc.

(c) Atom scattering method

In an atom scattering experiment, clusters are made to collide with light atoms, in a crossed-beam arrangement. The resultant momentum transfer during collision causes clusters of different sizes to scatter in different regions of velocity space, leading to separation of clusters of different size. Principle behind these experiments is that heavier clusters are deflected into smaller angles compared to the lighter clusters. Momentum transfer thus allows one to select the cluster of specific size and investigate its characteristics.[31, 32] For this purpose, generally light atoms with low collision energy are used as probe, to avoid collisional dissociation of cluster. In a typical experiment, clusters produced via supersonic expansion are subsequently crossed by another supersonic expansion consisting of pure expansion gas (He or other inert gas). The atoms, molecules, and clusters of the two supersonic jet collide and transfer momentum, as a result the clusters are scattered out of the first expansion beam at an angle proportional to their mass or size. Thus, clusters of different mass are spatially separated. A movable mass detector then quantifies the abundance of a given cluster size. However this process of mass selection has certain disadvantages like low concentration of mass selected cluster species and also collision can cause vibrational and rotational excitation of the cluster.

Another method which has been used for size selection of clusters is based on the time of flight method. This method is based on the dynamics of cluster formation in a supersonic expansion. This method makes use of the fact that during cluster nucleation and growth the time needed to form higher clusters is comparatively larger compared to formation of smaller clusters. Also during their directional movement under the influence of large pressure gradient these cluster experience velocity slip due to their different masses. Hence, in a pulsed supersonic beam, monomers such as the inert carrier gas and the constituent molecule vapours travel faster, while the cluster species are slightly decelerated according to their masses. Accordingly, in a gas pulse the different clusters are separated in time w.r.t the pulsed valve opening. The arrival distribution time of species with different masses is maximum at different time in the gas pulse, with respect to the nozzle trigger pulse. Thus,

spectroscopic features appearing in a given mass channel (timed by the ionization pulse) will have different intensity profiles with regard to the nozzle triggering pulse. However this method has limited utility as for larger cluster (higher masses) the difference in arrival time is insignificant. Large clusters form later in the expansion because they take more time to form and also because they travel more slowly

(d) Mass spectrometer

Mass spectrometers form an integral part of cluster research for mass analysis and mass selection. Also the intensity of the mass spectra of cluster ions carries information on the relative stability of the clusters. But as discussed above mass spectrometers are hardly used for characterization of size distribution of neutral clusters. On the contrary, as mass selection of cluster ion in a mass spectrometer is straightforward, they find use for production of size specific cluster ion beam, which can be reneutralized to produce monodispersed neutral clusters. Generally, negative ion clusters are neutralized by photodetachment, collisional detachment or charge exchange, while positive ion clusters are neutralized by resonant or non-resonant charge exchange.[33, 34]

Different types of mass spectrometer have found application in cluster related studies- for example Wien Filter, Magnetic sector instrument, ICR, Quadrupole, to name a few.[35]. However, Time of flight mass spectrometer (TOFMS) is the instrument of choice for cluster studies due to their unlimited upper mass range, higher speed and large ion transmission. Also it is inexpensive, easy to build and a complete mass spectrum can be recorded every few microseconds. In addition, it can be efficiently coupled to the pulsed cluster sources. Broadly TOFMS instrument can be divided into three parts, the ionization region, field free region and the detector region. In the ionization region, the clusters are ionized by a suitable ionization source (intense laser, electron beam, etc) the resultant cluster ions produced get separated according to their masses in the field free region and arrive at the detector at different time corresponding to their masses. Lighter fragment ions arrive early at the detector while heavier cluster ions arrive late at the detector.

(e) Optical methods

As clusters are microscopic objects, optical spectroscopic techniques, which have been widely exploited for characterization of molecular system, have also been successfully applied to clusters. Different interactions within a cluster (e.g. vander Waals, electrostatic, hydrogen bonding and charge transfer, etc.) can be understood through spectroscopic cluster shift. Similarly photoelectron spectroscopy and ZEKE-spectroscopy, which measure the kinetic energy of photo ejected electrons, have been used for obtaining details regarding the energy level structure of neutral clusters. Cluster energetics, structure and dynamics have been probed spectroscopically by microwave, Raman, infrared, visible, ultraviolet, one/two photon excitation, etc. techniques. In addition to common IR and UV direct absorption methods, several of their variants such as depletion spectroscopy, infrared photofragmentation spectroscopy, Laser induced fluorescence (LIF), Resonance enhanced multiphoton ionization (REMPI), Cavity Ring Down laser spectroscopy (CRDS) etc. have been successfully applied for obtaining the information regarding the electronic and structural properties of cluster, as a function of average cluster size.[36-40] In the following paragraph, we briefly describe the two sensitive techniques i.e. Ion dip IR spectroscopy and Cavity Ring Down Spectroscopy (CRDS).

(i) Ion dip IR spectroscopy: To know the structure of the complex, one needs to know the vibrational frequencies of the cluster. However, due to low concentration of the specific size cluster species, direct infrared absorption method does not work well. But as ions can be detected in small concentration due to high detection efficiency coupled with large amplification offered by ion detectors. Hence use is made of ion dip IR spectroscopy, which involves infrared absorption coupled with UV ionization of cluster, for detection (figure 5). Experimentally a UV laser at fixed resonant frequency is used to generate ions giving a constant signal. Since vibrational relaxation period for clusters upon IR excitation are longer as compared to the pulsed width of UV ionizing laser, the IR laser is fired just before (50-100 ns) the UV laser. No observable effect will be seen if the IR

laser is not in resonance with a vibrational transition. However, if the IR laser is in resonance with a transition, it populates the upper level, thus leaving lesser number of molecules in the ground state to be ionized by the subsequent UV laser. Thus, one observes a dip in the ion signal at vibrational frequencies that are in resonance with any of the transitions of the cluster.

(ii) Cavity Ring Down Spectroscopy (CRDS): In a typical ring down experiment, a short laser pulse is injected into a cavity formed by two highly reflective mirrors (reflectivity greater than 99.99%) containing the clusters whose spectra is to be measured. The reflective mirrors are separated by a distance, such that it is larger than the laser coherence length. Under suitable conditions, the light pulse bounces back and forth in the cavity. Light coming out of the cavity is monitored by a suitable detector. If the detector has a fast enough response time then it will see individual laser pulses leaking out of the cavity separated in time by the cavity round trip time and with an exponentially decreasing intensity.

By measuring the decay constant of the exponentially decreasing light intensity, the absorption of the low concentration species present in the cavity can be measured. CRDS has been used to study a variety of metal clusters and metal containing molecules formed by laser ablation in supersonic beams. The hydroxyl (OH) stretching bands of a variety of hydrogen oxide $(H_2O)_n$ and deuterium oxide $(D_2O)_n$ clusters were measured for n = 2-6 with a resolution of 0.04 cm^{-1}.[41] A rotationally resolved spectra of water dimer $(H_2O)_2$, was measured which provided invaluable information for the development of an accurate water pair potential.

A combination of these above mentioned techniques have been widely used for understanding the properties of mass selected clusters. A wealth of information, such as the ionization potential, electron affinity, dissociation energies, gap between highest occupied molecular orbital (HOMO) and lowest unoccupied molecular orbital (LUMO), reactivity etc. have been obtained for characterizing and to study the evolution of their properties with size and composition.

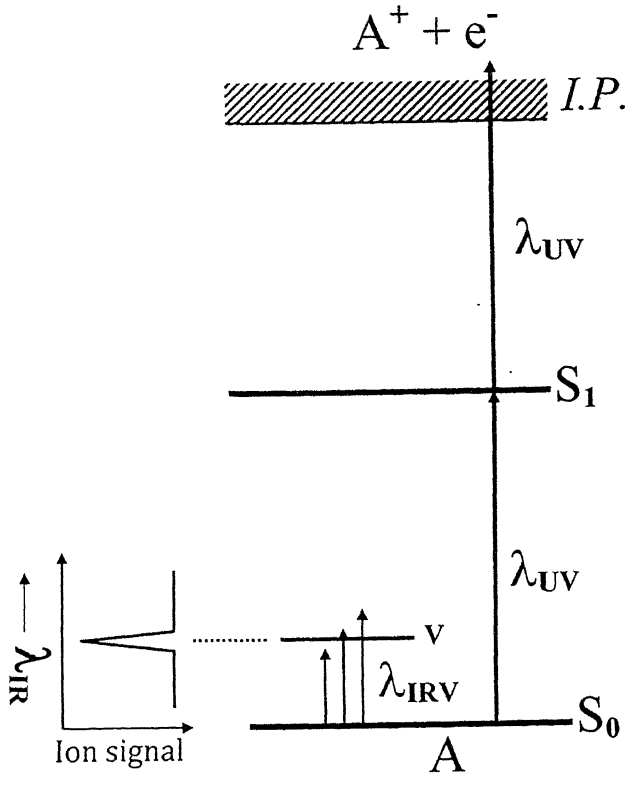


Fig. 5– In IR ion-dip spectroscopy, a molecule/cluster is resonantly/non-resonantly ionized by an UV laser at a fixed wavelength and simultaneously an IR laser is scanned to probe the vibrational bands of the molecule/cluster. Upon resonance with the vibrational level a sharp decrease is observed in the ion signal due to depopulation of molecules/cluster from the ground electronic state.

Photochemistry of clusters and comparison with their monomer counterpart

Clusters, like their monomer counterpart (molecules) have also been subjected to optical studies for investigation of cluster structures and dynamics, as well as to realize the effect of cluster formation on the photophysical properties of molecules. The variation in the photochemical behaviour of clusters and that of its corresponding molecular analogue in gas phase mainly depends on the properties of the initial state which participate in photoexcitation process. Various physicochemical processes starting from excited electronic-state such

as vibrational relaxation, vibrational predissociation, electron transfer, proton transfer, radical additions etc. are initiated in clusters upon absorption of photons. These reactions have been investigated by different experimental approaches and detection techniques discussed above. These studies are essential for understanding the influence which solvation, degree of aggregation and the proximity of solute complexes have on the reaction and properties of molecular species.[42]. It has been illustrated that cluster formation lowers the ionization threshold as well as the electronic origins relative to those of the monomer, which in turn can introduce profound changes in the photodynamic behaviour of molecules that are incorporated into the cluster. For example in water-ozone complex (O_3:H_2O) there is a red shift in the absorption of ozone and its absorption cross-section at 355 nm increases by about two orders of magnitude compared to that of the isolated ozone molecule.[43]. Also, in certain cases, vander Waals clusters have been shown to exhibit concerted photochemistry, where new chemical channels open up which do not exist for the isolated molecule. For example, dimers of OCS and CS_2 generate S_2 as a photofragment, while O_2-O_2 complex gives rise to O_3+O at excitation wavelengths, well below the dissociation threshold of their isolated counterpart.[44] Similarly, water clusters are of interest for several reasons (atmospheric and biological importance). Water clusters have been predicted to be present in atmosphere, and presence of water dimers has been confirmed recently. These clusters are assumed to be important for oxidation of SO_3 to H_2SO_4 and to HO_2 radicals in the atmosphere. Also water clusters can provide important leads in understanding nucleation process which results in transformation of water vapours upon condensation, into water droplets resulting in rain as well as formation of snow and ice. They have also been found to congregate in the confined cavities of proteins and other biomolecules.[45, 46] As water is an universal protic solvent, a large number of studies have also been carried out on complexes of different molecules with water clusters. These studies are motivated with the desire to understand solute-solvent interactions and the influence of H-bonding interaction within water molecules on the properties of solute.[47, 48]

Clusters have also been subjected to intense lasers pulses, to understand their multiphoton excitation behaviour. Following excitation/ionization, the clusters exhibit different intracluster reactions, which depend on degree of aggregation of the cluster as well as the mechanism of energy dissipation and transfer. For example, on multiphoton ionization, clusters of acetone undergo ion-molecular reaction leading to formation of solvated acetone cluster ions i.e. $[(CH_3)_2CO]_mH^+$, $[(CH_3)_2CO]_mC_2H_3O^+$ and $[(CH_3)_2CO]_mCH_3^+$ ions. These cluster fragments ions are produced either from ionization of an excited species formed by dissociation of intact cluster precursor or by dissociation, after the intact cluster species has been ionized. Previous, pump probe studies carried out on these acetone clusters suggest that for monomers and dimers dissociation of neutral clusters followed by ionization of cluster fragment is the dominant mechanism. While for higher clusters ionization of the cluster followed by its fragmentation is the dominant mechanism.[49, 50]

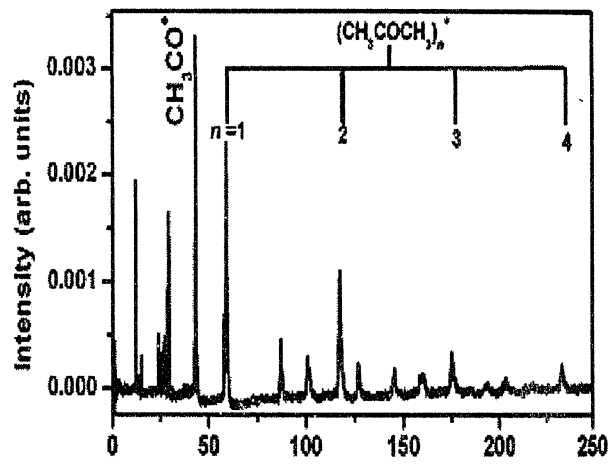


Fig. 6– Time of flight mass spectra of acetone clusters subjected to 532 nm laser pulses.

(a) Cage effect in the photolysis of methyl iodide clusters

We have investigated photochemistry of methyl iodide monomer and cluster at 266 nm.[51, 52] The well known $\tilde{A}$ state of CH_3I, corresponding to the excitation of a non-bonding 5pπ iodine electron to the σ^* antibonding C-I orbital, lies in 330 – 210 nm range. Excitation to $\tilde{A}$ state results in rapid direct

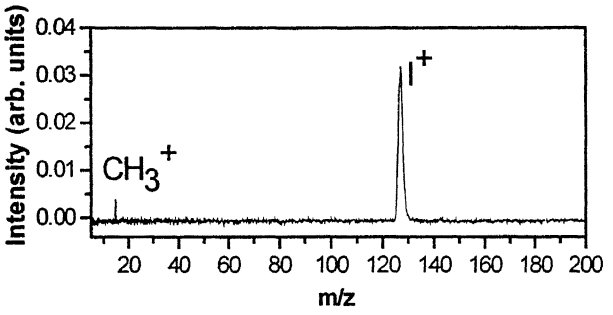
dissociation producing CH_3 radical and iodine atoms on a time scale of ~100 fs.[53] In view of the dissociative nature of $\tilde{A}$ state, at 266 nm CH_3I should undergo fast dissociation leading to absence of molecular ion in the time of flight spectra. This is apparent from the time of flight mass spectra of methyl iodide monomer recorded after multiphoton excitation at 266 nm (fig. 7(a)). In our experiment we have used a nanosecond laser and the process of one photon dissociation via $\tilde{A}$ state dominates over multiphoton excitation. This results in dissociation of methyl iodide monomer into CH_3 and I fragments, which are subsequently ionized. Thus resulting in absence of CH_3I^+ ion in the MPI spectrum. In another experiment to understand the effect of solvation on the photochemical dynamics of the dissociative $\tilde{A}$ state of CH_3I, we have subjected methyl iodide clusters to 266 nm laser pulse. Figure 7(b) shows a typical time of flight spectrum for a methyl iodide cluster beam irradiated with mildly focused 266 nm laser radiation. The ion peaks observed in the mass spectra correspond to ions of C^+, CH_3^+, I^+, CH_3I^+, $(CH_3)_2I^+$, I_2^+, $CH_3I_2^+$ and $(CH_3I)_2^+$. On comparison of time of flight mass spectra of methyl iodide cluster with that of methyl iodide monomer recorded at 266 nm, we find that additional ion peaks for CH_3I^+, $(CH_3)_2I^+$, I_2^+, $CH_3I_2^+$ and $(CH_3I)_2^+$ ions are observed in the mass spectra of CH_3I clusters. In case of CH_3I monomer the absence of molecular ion peak was explained on the basis of fast dissociation of CH_3I molecule after single photon excitation to repulsive $\tilde{A}$ state. Hence, in case of clusters also if the mechanism of photo-excitation process was similar, the molecular ions should have been absent. However, observation of CH_3I^+, $(CH_3)_2I^+$, $CH_3I_2^+$ and $(CH_3I)_2^+$ in the mass spectra obtained on multiphoton ionization of methyl iodide clusters suggests that for large clusters caging can occur, which can result in stabilization of the valence $\tilde{A}$ state due to intermolecular interactions.[54] Here cluster caging refers to the influence of the surrounding medium on the behaviour of reaction intermediates or products. Depending on the system, the surrounding molecules that constitute the cluster cage may prevent the mutual separation of the product and lead to recombination or else delay the separation of the products and influence their energy distribution.

In addition, observation of I_2^+ ion in the mass spectra of methyl iodide cluster indicates that cluster undergo intramolecular reactions since the monomer MPI spectra of CH_3I shows peaks for CH_3^+ and I^+ only which are the primary photoproducts at 266 nm. Several previous studies, dealing with $\tilde{A}$ state assisted multiphoton ionization of CH_3I clusters, have reported the observation of I_2^+ ion in the mass spectra. However, the mechanism of I_2^+ formation is still a matter of controversy, though it is universally accepted that the lower clusters $[(CH_3I)_n$, where n = 2-3] are responsible for presence of I_2^+ ion in the mass spectra. To explain the mechanism of I_2^+ ion formation some of the researchers have proposed that these ions are produced via concerted photodissociation of ionized cluster $(CH_3I)_2^+$,[55, 56] while others are of the opinion that formation of neutral I_2 (via concerted or sequential photolytic mechanism within the cluster) followed by its ionization is the most probable mechanism.[54]

(b) Rearrangement and elimination in cluster fragment ions

In another experiment, clusters of $(CH_3SCH_3)_n$ and $(CH_3SSCH_3)_n$ were subjected to laser pulses of gigawatt intensity at 355. Figure 8. shows time of flight mass spectrum of CH_3SCH_3 clusters at 355 nm under laser intensity of ~ 3.5 x 10^9 W/cm^2. The lower mass range of the spectra is shown in figure 8(a). While the higher mass range is shown in figure 8(b), for the sake of clarity. In figure 8(a). the ion signals at m/z = 12, 13, 14, 15, 32, 35, 45, 46, 47, 61 and 62 were assigned to C^+, CH^+, CH_2^+, CH_3^+, S^+, SH_3^+, HCS^+, CH_2S^+, CH_3S^+, $CH_3SCH_2^+$ and $CH_3SCH_3^+$. Peaks in the C_2H_n region are indicative of the existence of C–C bond for some of the ionized species. While in figure 8(b). higher cluster fragment ions such as $CH_3(CH_3SCH_3)^+$, $CH_3S(CH_3SCH_3)^+$, $(CH_3SCH_3)_2^+$ and $CH_3(CH_3SCH_3)_3^+$ have been observed. It is worth noting that the most intense ion peak in the mass spectra is for HCS^+, which is a secondary fragmentation product and is formed from unimolecular elimination of H_2 from CH_3S^+.[57] On comparing the fragmentation mass spectra of DMS monomer at 355 nm, which has been reported earlier from our lab[58], we find that the fragmentation mass spectra of $(CH_3SCH_3)_n$ are not much different except

that in the higher mass range cluster fragment ions are also observed. But it is worth noting that the ion signal for SH_3^+ ion and $CH_3SCH_2^+$ ion are much more prominent in the present study. In a previous study, it was proposed that SH_3^+ ions originate from $CH_3SCH_2^+$ ion via a cyclic intermediate state (Scheme I.). The results suggest that abundance of $CH_3SCH_2^+$ and the subsequent cyclization reaction leading to SH_3^+ is facilitated in clusters.



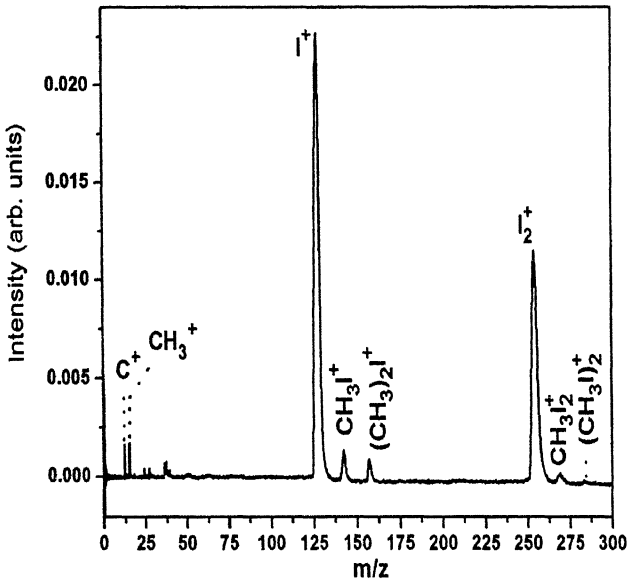
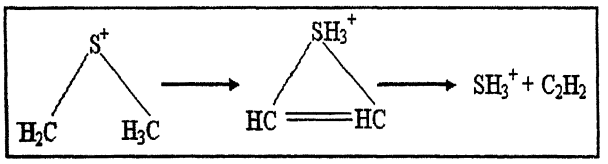


Fig. 7– Time of flight mass spectra of CH_3I monomer and clusters subjected to 266 nm laser pulses.



Scheme I.

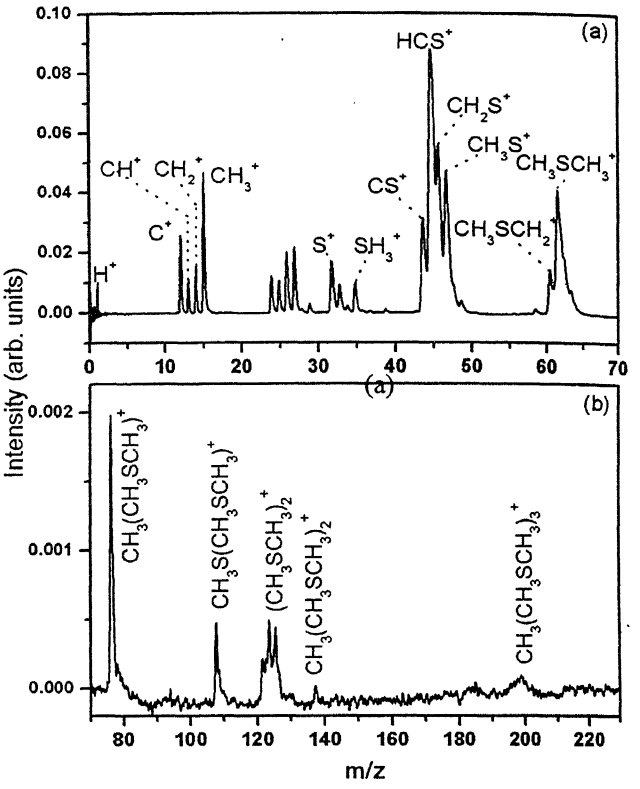


Fig. 8– Time of flight mass spectra of dimethy sulphide clusters subjected to 355 nm laser pulses (a) m/z=1-70 and (b) /z= 70-230.

(c) Coulomb explosion studies in vander Waal's clusters

Clusters have high local density of electrons and atoms and this property helps them to interact efficiently with laser radiation. When these clusters were made to interact with laser pulses of intensity 10^{14}-10^{16} W/cm^2 a novel phenomenon was observed i.e. multiply charged ions of the constituent atoms with very high kinetic energy were observed. In addition to this, generation of highly energetic electrons, emission of X-rays and even neutrons were reported. This phenomenon is termed Coulomb explosion, since it arises from very strong (almost explosive) repulsion of highly charged cluster matter generated as a result of severe ionization (removal of several electrons) of cluster constituents. This phenomenon has suggested utilization of clusters as precursors for generation, upon Coulomb explosion of highly charged clusters, of multiply charged energetic ions and electrons, enhanced emission of

X-rays, and even neutrons, for several applications. Study of these laser-cluster interaction processes are motivated by the desire to generate higher order harmonics[59], energetic electrons[60], multiply charged ions[61] and even neutrons.[62]

Summary/conclusions

We have shown that the interaction of light with clusters may lead to observation of different physico-chemical phenomena. Clusters exhibit altogether different photochemistry, as new photochemical reaction channels open up, which are not observed for the molecules. Optical response of cluster provides interesting information on clusters, in particular because of its size dependence. The case of irradiations by intense lasers opens door to a new challenging phenomena, such as the production of highly energetic species (photons, electrons ions). In this domain of physics, the interaction process between the cluster and the laser is so strong that it leads to a severe ionization and extensive fragmentation.

Acknowledgement

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Status of the endemic plant *Hypericum gaitii* (Hypericaceae) in Similipal biosphere reserve of Orissa: A need for conservation

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Abstract

Hypericum gaitii, an endemic shrub to Orissa, India, which is located in Similipal Biosphere Reserve, appears to be restricted to five extant subpopulations. Intensive surveys are required in order to establish whether there are any other extant subpopulations exist in other part of Orissa, and the presently known subpopulations require habitat monitoring and continuous protection.

Keywords: critically endangered, endemic, hypericaceae, *Hypericum gaitii*, Orissa, similipal biosphere reserve

India is one of the 12 mega-biodiversity countries in the world including two hotspots[1]. It occupies 2.4% of world's land area and contributes nearly 8% of world's biodiversity. However, conservative estimates suggest that at least 10% of India's recorded wild flora and 20% of its mammals are on the threatened list[2]. About 45,000 plant species are reported to occur in India, representing 11% of the known world flora. Among them, 33% flowering plant and 29% of the total Indian flora are endemic which contributes to the rich plant diversity in the country[3].

Hypericum is a genus of about 400 species of flowering plants in world belongs to family Hypericaceae. They are mostly distributed in temperate regions of the world, missing only from tropical lowlands, deserts and arctic regions. Out of 400 species of this genus, about 29 species occur in India[4]. The type species of this genus, *Hypericum gaitii* is distributed in Peninsular India and found only in parts of Jharkhand and Orissa state. Haines collected and described the species for first time from northern Orissa[5]. Unfortunately, so far the precise locality in Jharkhand state was not recorded. In Flora of Orissa, this species is reported from Similipal Biosphere Reserve (SBR), Orissa[6]. Many taxonomical works has been done in Orissa but no one reported this species from any other parts of Orissa state except Haines, Saxena and Brahmam. There is also not much information available on this species. Earlier, Haines, Saxena and Brahmam mentioned that this endemic species is threatened by several factors and in verge of extinction[7]. Out of 28 endemic plants found in Orissa, *Hypericum gaitii* is one of them, having very narrow range of distribution. Nevertheless, there has been no research on the species since last one decade. After gap of a decade, the authors were able to relocate this species from SBR in Mayurbhanj district of Orissa. Here, a few subpopulations of this species are surviving in fragmented forest patches of moist peninsular high level Sal (*Shorea robusta*) forest. However, lack of information on population size and distribution in Orissa make it difficult to access the effect of past management regulations and to determine its present conservation status.

Study area

The study was conducted in the Similipal Biosphere Reserve (SBR) of Mayurbhanj district, Orissa. SBR is the only biosphere reserve of the Eastern Ghats, situated between 21° 28' to 22° 08' N latitude and 86° 03' to 86° 37' E longitude (Fig. 1). It has been declared as a wildlife sanctuary comprising a forest area of 2,200 sq. km during 1979.

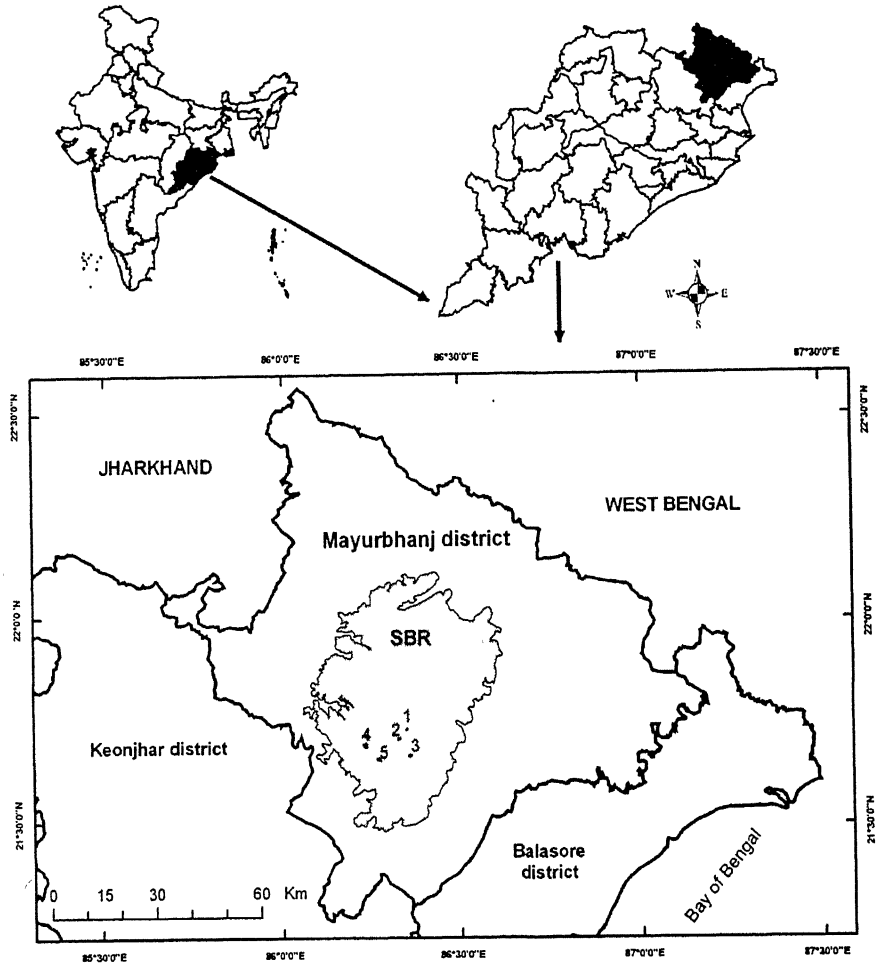

Fig. 1– The location of the five subpopulations (1-5) of *H. gaitii* on SBR, Orissa. (1) Jenabil East with 70 individuals; (2) Jenabil North with 115 individuals; (3) Jenabil South with 184 individuals; (4) Upper Barkmuda with 245 individuals; (5) Deosthali with 156 individuals.

Government of India declared Similipal as a biosphere reserve due to its rich biodiversity and natural heritage on 22nd June 1994. The Eastern Ghats are delimited in the north by Similipal hill ranges of Orissa. Geological formation of the region consists of sub-metamorphic sandstones and quartzite haematites[8]. The soil is reddish in colour and loam to sandy loam in texture. The soil is slightly acidic in nature with P^H ranging from 5.23 to 6.52 and average monthly soil moisture content[9] varies from 18.13 to 40.25%. The climate of the area is monsoonal type and rain occurs due to northeast monsoon. The average annual rainfall is up to 500 mm, and is largely restricted to the period from July to October. The mean maximum temperature varies from 16 °C (December) to 35 °C (June) and mean minimum temperature from 6 °C (January) to 22 °C (June). The natural vegetation is mostly moist deciduous type[10], as Sal is one of the dominant species. There are 1076 plants recorded from the area including 60 species of ferns, 92 species of orchids and two gymnosperms[11-12].

Extensive surveys were undertaken during 2004 to 2006 in the study area for the purpose of collection of plants as part of the Eastern Ghats biodiversity project. Phytosociological data were collected in 10 x 10 m quadrants which were

Table 1- Summary of *Hypericum gaitii* sites in Similipal Biosphere Reserve, Orissa

Site	Latitude	Longitude	Elevation range (m)	Aspect	Individuals
Jenabil East	86° 20' 56" E	21° 43' 58" N	550-630	N-NE	70
Jenabil North	86° 19' 38" E	21° 42' 30" N	580-660	NE	115
Jenabil South	86° 21' 27" E	21° 40' 04" N	500-580	NE	184
Upper Barkmuda	86° 16' 08" E	21° 39' 33" N	900-1,000	SE	245
Deosthali	86° 13' 52" E	21° 41' 36" N	840-900	SE	156

Fig. 2- Endemic plant *Hypericum gaitii* Haines

systematically surveyed for all shrub and herb species. The collected species were identified with the help of the Flora of Orissa and specimens preserved in the herbarium of Kakatiya University (KUH), Warangal (Andhra Pradesh). As the occurrence of the species was not so frequent, we collected few specimens for identification during our 5-6 km walk along the narrow forest road of SBR (Fig. 2). Global Positioning System (GPS) points were collected wherever the species was found (Fig. 1). The site locations with latitude, longitude, elevation, aspect and populations are given in Table 1.

Results and Discussion

The species is mostly found in the fringe areas of moist Sal vegetation, which are generally affected by frost in the winter season[11]. The area of occurrence of this species is marked with the help of GPS taken in the field in order to point out locations and declaring 'zones for conservation' of the species in future. During our survey, we were able to collect this species in its flowering and fruiting period. The other associated species found along with *H. gaitii* are given in Table 2. In surveys for *H. gaitii* carried out in 2004 and 2006, we found only five subpopulations, in the north-eastern part of SBR. A count of each subpopulation indicated that the total population is *c.* 770 individuals, with subpopulations varying in size from 70 to 245. The species are generally found in north-east and south-east aspect, at altitudes of 550-1,000 m.

Proper and adequate information regarding the taxa, its ecological requirements and population dynamics are essential for the conservation and

Table 2– Associated plant species found along with *Hypericum gaitii*

Plant species	Family	Habit
Cassia hirsuta L.	Caesalpiniaceae	Shrub
Chromolaena odorata (L.) R.King & H.Robins.	Asteraceae	Shrub
Desmodium pulchellum (L.) Benth	Papilionaceae	Shrub
Dicliptera bupleuroides Nees	Acanthaceae	Herb
Flemingia chappar Buch.-Ham. ex Benth	Papilionaceae	Shrub
Galactia tenuiflora (Klein ex Willd.) Wight & Arn.	Papilionaceae	Climber
Indigofera cassioides Rottl. ex DC.	Papilionaceae	Shrub
Melastoma malabathricum L.	Melastomaceae	Shrub
Osbeckia stellata Buch.-Ham.ex Ker-Gawl.	Melastomaceae	Herb
Shorea robusta Gaertn.f.	Dipterocarpaceae	Tree

preservation of the species. This may be the main reason why this species is not listed in the IUCN Red List category, Red Data Book of Indian flowering plants and the scheduled category of plants in the Indian Wild Life Protection Act 1972. This species is regarded as a threatened and important because (1) it is endemic to Peninsular India, i.e. specific to Orissa, with only five subpopulations currently known that comprise a total of *c.* 770 individuals found only in a restricted area, (2) these subpopulations are only found along with the moist Peninsular high level Sal forest, which is not found anywhere in Orissa, (3) the area of extent of the subpopulations are less than 10 km^2, (4) there is evidence that the species is being collected and used in treatment of skin diseases by the local tribal people of the surrounding areas[13], and (5) the typical habitat of the species has been markedly reduced by past human influence. Based on above information it should be categorized as Critically Endangered on the IUCN Red List[14], based on criteria B2a+b i.e. with an area of occupancy[15] of <10 km^2 (B2), severely fragmented (a), and with a continuing projected decline (b) in extent of habitat.

Conservation measures

The major threat to *H. gaitii* is habitat loss due to high anthropogenic pressures. In Orissa, tribal people are mostly depending on forest for fuelwood and other non-timber forest products (NTFP) for their livelihood. These species are found on the edge of the forest and nearer to agricultural fields. This poses a major threat to the existence of the species because of trampling and cutting by local people. Since we could locate the species growing only on the fringe areas of forest, extra care should be taken regarding this aspect in order to conserve this ecofragile system. Deforestation, regular grazing by cattle from near by habitations and possibly burning are also bring natural catastrophe in this area. Another major threat for the species is from tourism. Some of the subpopulations were marked just at the side of the tourist road of SBR. The population of this taxon in this area is not viable, because the population size is insufficient for the successful existence of a species according to the conservation rules. The narrow distributional range, low degree of habitat protection, microhabitat specificity, nonviable population, etc. is bringing this species closer to extinction. Both *in-situ* and *ex-situ* conservation strategy should be adopted to save this remarkable species from the verge of extinction. Public awareness and conservation education programmes should be intensified so as to effectively conserve such botanically interesting species. Any natural disaster like diseases, pest attack, etc. may eventually wipe out the remaining populations of this endemic and rare species from the world. Therefore, intensive botanical surveys are required

in other parts of Orissa in order to determine whether there are any other extant populations of the species, and the presently known subpopulations require long-term monitoring and continuous protection.

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Entity relationship model to illustrate molecular evolution

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Abstract

The entity relationship (ER) modeling grammar is a tool for the illustration and effective communication of conceptual designs of commercial/business systems. Hitherto, ER models are being used in the computer science and information systems disciplines. The discipline of biology involves large amount of data representing several facets including the physiological and phylogenetic aspects of organisms. In this paper, the intricacies involved in the process of molecular evolution have been modeled using the ER modeling grammar. The principal components of molecular networks elucidated in a vignette are classified as the entity types. Each entity type is defined by one or more unique identifiers and other optional and/or mandatory attributes. Relationships among the entity types are described through the cardinality ratio and participation constraints. Microsoft/Visio, a software engineering tool is used to develop the ER diagram.

Keywords: conceptual design, ER modeling grammar, molecular evolution

Introduction

The subject of biology is limitless. Biology has come into limelight through the evolutionary processes. Charles Darwin[1] introduced biology through the concept of Natural Selection elucidated in his book entitled "The Origin of Species". Darwin explicitly elaborated on the Natural Selection favouring survival and adaptability of organisms in their respective habitat[2]. In pursuance of the same, the whole period of 20th century was engaged in the collection and compiling of biological data[3]. Ernst Mayr[4] vividly elucidated the importance and use of biological data relating to species, their habitat, pathogenicity, control of population and ultimately the equilibrium within the biodiversity. Of late, the curator centers of biological data such as NCBI, EBI, EMBL, EXPASY, etc. have arisen. The urge for molecular information started since the time the unfolding of the structure of DNA[5] and its elucidation as the core biological informational molecule came into vogue[6]. In consonance with the development of tools and techniques in genomics, there has been an enormous accumulation of data which needs to be organised and compiled to comprehend the hidden principles of evolution of life. The uniqueness of biological information is that attributes of biological systems will never become obsolete, Instead, the depth of information of a biological attribute incessantly keeps growing through molecular evidences and genomic mining of representative members of biodiversity, Thus, there is an enormous scope in biology to effectively simplify the communication by depicting complex biological systems and data in organised databases. In this regard, data modeling and database design techniques developed in the Computer Science (CS) and Information Systems (IS) disciplines could be of value to the biologist.

The Entity-Relationship (ER) modeling grammar as a conceptual modeling tool was proposed by Chen[7] For over three decades, research in conceptual data modeling has embellished the constructs and rules of ER modeling grammar to meet the needs of the growing complexity of business and commercial domains, ER model was

also chosen by the American National Standards Institute (ANSI) in 1988 as the standard model for information resource directory systems (IRDS)[8], ER modeling grammar is an effective tool for communicating technical information and widely used in the CS and IS domains to capture complex data structures inherent in the business management and commercial application domains[9].

In the present endeavour, an attempt has been made to adopt the ER modeling technique to capture and communicate the intricate processes involved in molecular evolution. The present article is an elucidation of an approach that models the principles of molecular evolution as database.

Methodology

The ER modeling language employs a graphical technique. The three building blocks of an ER model are: Attributes, entities and relationships. To begin with concepts of molecular evolutionary processes are described in a vignette. The development of an ER diagram often begins by scrutinizing processes described in the vignette to extract possible entity types, attributes, and relationships among entity types. In brief, a real world object is conceptualized as an entity. The object type can have many properties. Correspondingly, an entity type is composed of attributes. The association between the entities is referred to as relationship. Attributes have several characteristics (e.g., Name, Type, Classification, Role). An entity type is a collection of related attributes. Relationships occur between entity types. Relationships are defined by cardinality ratio (m:n, 1 :m, 1:1) and participation constraints (total or partial participation). Umanath and Scamell[9] suggest that the conceptual modeling is a heuristic process, as opposed to a scientific process, and is therefore intuitive and iterative in nature passing through several punctuated equilibriums depending on the number of iterations in modeling dictated by the complexity of the application domain. The ER diagram notation is shown in Fig. 1. Initially, the discernable entity types are listed from the vignette and their attributes are identified based on apparent commonalities among them (Tables 1, 2 and 3). The graphical tool, Microsoft/Visio, was used to develop the ER diagram for the molecular evolution as shown in Fig. 2.

Vignette describing molecular evolution

The bewildering biodiversity around us is the ultimate product of organic evolution. Phenotypic diversity and genotypic diversity in a population result from evolution. There are morphological, behavioural and anatomical variants within the *phenotypic diversity.* The *genotype diversity* involves the variation in the chromosomes and DNA. The variation in the specified sequences of nucleotides in the Genomic DNA is called DNA polymorphism. The genetic map and physical map of chromosomes authenticate genetic diversity in evolution. The prevailing polymorphic DNA sequences are either due to single nucleotide polymorphism or restriction fragment length polymorphism or recombination. The same would . have been the root cause for the existing biodiversity around us.

The gene is a hereditary unit. It is also a functional unit of DNA. The underlying force for evolution is concealed in genes. The alteration within the linear sequence of genes is the primary cause for organic evolution. These alterations led to the emergence of several gene families. Thus, *new incipient genes* find a place within the organic world. Genes undergo *mutations.* They cause alterations in protein sequences (phenotype diversity). Thus, genes are informational molecules from which working molecules transcribe and translate as phenotypes. Single or many nucleotides, either by point or frame shift substitutions may cause genes to mutate. A gene undergoes mutation due to one or more *nucleotide substitutions.* Point mutations are due to transitions or transversions of purines and pyrimidine bases.

Meiosis is a reductional cell division. The germinal cells undergo spermatogenesis in testes and oogenesis in ovary. They are the reduction divisions occurring in germinal cells. At times, during the reduction division due to *unequal crossing over,* genes either duplicate or inter exchange their exons through chromosome loops or miss in gametes. However, *genes do not depend on meiosis for their revision.* Thus, there is an enormous scope for bringing about the variation within the organic world. However, the velocity of organic evolution is kept at a low pace due to several *synonymous mutations* and *genetic recombinations.*

Table 1– Entity types involved in the description of molecular evolution.

S.N.	Entity type	Attribute	Optionality	Role	Classification
1	Evolution	Phenotype:•	Mandatory	Unique Identifier	Composite
		Behavioural	Optional	Key attribute	Atomic
		Morphological	Optional	Key attribute	Atomic
		Anatomical	Optional	Key attribute	Atomic
		Genotype:	Mandatory	Unique identifier	Composite
		Genetic map	Optional	Key attribute	Atomic
		Physical map	Optional	Key attribute	Atomic
		SNP,RFLP	optional	Key attribute	Atomic
2	Gene	Functional unit	Mandatory	Unique identifier	Atomic
		Unit of Heredity	Mandatory	Unique identifier	Atomic
		Revision:	Mandatory	Unique Identifier	Composite
		Retain	Optional	Key attribute	Atomic
		Replace	Optional	Key attribute	Atomic
3	Nucleotide	Point Mutation:	Mandatory	Unique Identifier	Composite
	Substitution	Transitions	Optional	Key attribute	Atomic
		T ransversions	Optional	Key attribute	Atomic
		Frame shift mutations	Optional	Non-key attribute	Atomic
4	Exon	SINE	Mandatory	Unique identifier	Atomic
	Shuffling	Transposons	Optional	Non-Key attribute	Atomic
5	Retrotrans	Reverse Transcriptase	Mandatory	Unique identifier	Atomic
	position	mRNA	Mandatory	Unique identifier	Atomic
6	Meiosis	Reduction division:	Mandatory	Unique identifier	Composite
		Spermatogenesis	Mandatory	Key attribute	Atomic
		Oogenesis	Mandatory	Key attribute	Atomic
		Unequal Crossing over	Mandatory	Unique attribute	Atomic

Table 2– The relationships shown below are between the entity types indicated in Table 1 describing molecular evolution.

S.N.	Relationship type	Degree	Entity type	Cardinality ratios	Participation Constraint
1	Deleterious mutations	Binary	Gene	1 : n	Mandatory
			Evolution		Mandatory
2	Neutral substitution	Binary	Gene	m : n	Optional
			Evolution		Mandatory
3	Neutral substitution	Binary	Gene	m : n	Optional
			Evolution		Mandatory
4	Loss / Gain of sequences	Binary	Gene	m : n	Optional
			Nucleotide substitution		Mandatory
5	Make a copy	Binary	Gene	n : m	Mandatory
			Retrotransposition.		Mandatory
6	Duplicate /Miss	Binary	Gene	1 : m	Optional
			Meiosis		Mandatory

Table 3– The Participation constraints and cardinality relationships.

S.No.		Participation constraints
1.	•	Deleterious mutations in one gene lead to several evolutionary changes.
	•	Several evolutionary changes have been due to deleterious mutations within a single gene.
	•	Many neutral substitutions of nucleotides in genes may not necessarily cause the evolution, but acknowledge evolution.
	•	The evolutionary changes are due to the neutral substitutions of nucleotides.
	•	Semantic integrity constraint: Deleterious mutations and neutral substitutions are mutually inclusivefor evolution to take place.
2.	•	Nucleotide substitution may cause the revision of a gene.
	•	Gene undergoes revision due to one or many nucleotide substitution.
3	•	Exon shuffling causes the loss / gain of nucleotide sequences in many genes.
	•	The loss / gain of sequences in a gene need not necessarily be due to exon shuffling.
	•	A gene requires at least one retrotranspoistion to make its own copy. Many units of retrotransposition can make multiple copies of genes.
	•	One gene can get copied by one or more retrotransposition.
5	•	Meiosis can duplicate / miss one or more genes.
	•	Genes do not require meiosis for their duplication/missing.

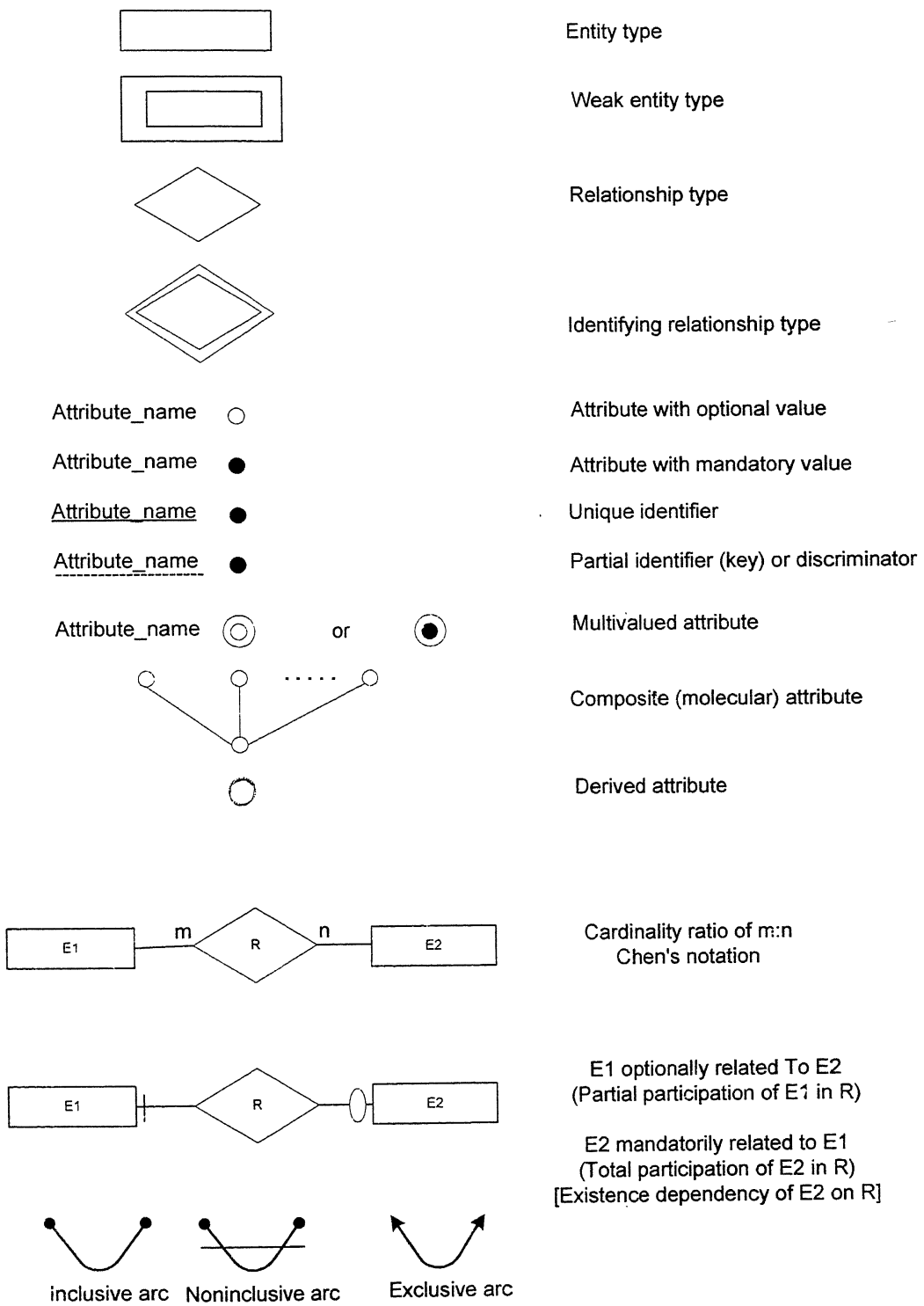

Fig. 1– Summary of ER diagram notation.

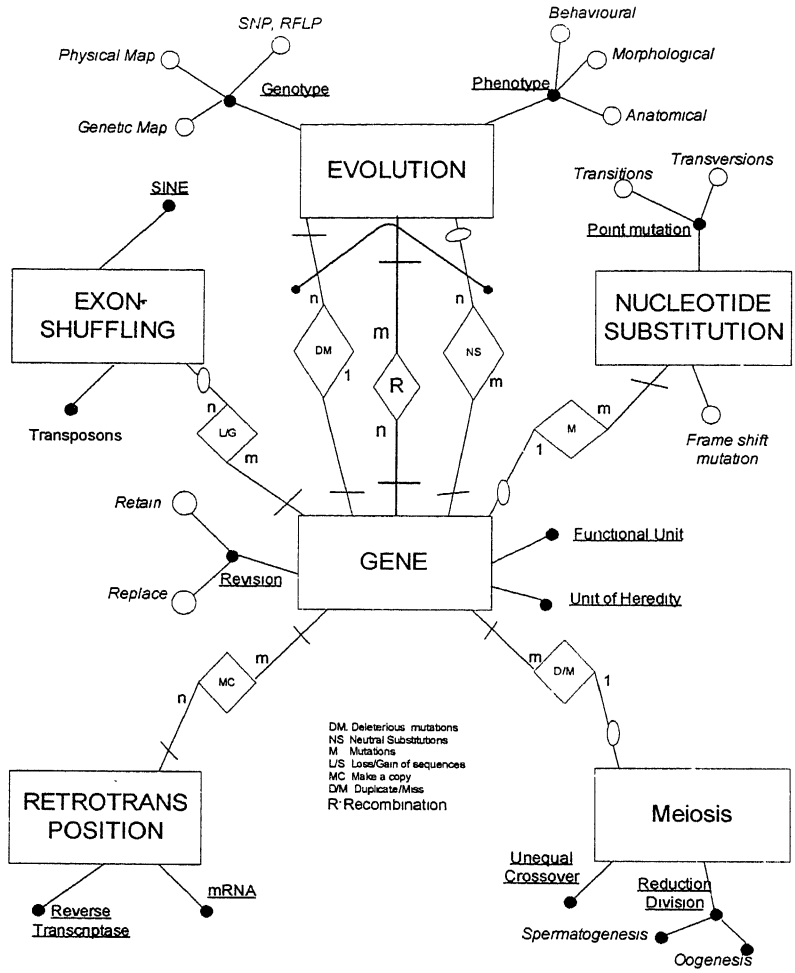

Fig. 2– ER diagram illustrating molecular evolution.

Yet, another process called *retrotransposition* which utilizes reverse transcriptase enzyme of remnant retroviruses (infected and integrated in the host genome once upon a time) to make another copy of a gene by assembling a natural cDNA using mRNA as a template. Thus, a gene requires at least one retrotransposition to make its own copy and many retrotranspositions can make several copies of genes. Alternatively, one gene can get copied by one or more transpositions. The *exon shuffling* by the so-called jumping machinery such as transposons and small interfering nuclear elements make a portion of a gene called exon to shuffle. This process makes the gene either to lose a portion of its frame or gain with unwanted frame. Moreover, the gene sequence need not necessarily be replaced due to exon shuffling. Therefore, the gene is the target for all the molecular mechanics. The gene may either be positively retained with tolerable (synonymous) substitutions or replaced with deleterious (non-synonymous) substitutions. Consequently, the launching of a new (revised) version of a gene takes place in a population subjecting for the test of Natural Selection that may be either advantageous or disadvantageous or neutral.

Conclusion

Thus, the generated ER diagram (Fig. 2) illustrates the principles and mechanics (database) of molecular evolution, the hallmark of organic evolution. In this ER diagram the adopted entity types' viz. Evolution, Gene, Nucleotide substitution, Exon shuffling, Retrotransposition and Meiosis are

distinctly identified from the vignette. The attributes are assigned to each one of the entity types viz. composite attribute, unique identifier, mandatory attributes and optional attributes. The cardinality ratios and participation constraints between entity types shown in the ER diagram clarify the intricate facets of molecular evolution. The diversified phenotypes and genotypes as noticed in the vast biodiversity are bestowed to us to contemplate on the pros and cons and intricacies of life bearing organisms.

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Aconitine alkaloids from tubers of *Aconitum heterophyllum* and *A. balfourii*: Critically endangered medicinal herbs of Indian Central Himalaya

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Abstract

Aconitum heterophyllum Wall and *A. balfourii* Stapf (Ranunculaceae), important medicinal herbs of the alpine and sub alpine belts of the Himalayan region are source of major diterpenoid alkaloids, namely aconitine and pseudoaconitine. Quantification of these alkaloids was carried out in tubers collected from higher altitudes in Kumaun and Garhwal regions of India, following column, thin layer and high performance liquid chromatography with an aim of identifying elites. The aconitine levels in different populations of *A. heterophyllum* varied from 0.13-0.75% (dry weight basis); maximum and minimum levels were detected in tubers from Phurkia (3260 m altitude) and Kafni (3400 m), respectively. In *A. balfourii* the amount of aconitine and pseudoaconitine also varied and ranged from 0.13-0.83% and 0.06-0.62%, respectively. Highest level of pseudoaconitine (0.62%) was recorded from Phurkia Bugyal (3430 m) while the lowest and about ten-fold less value (0.06%) was recorded in tubers from Kafni population (3400 m); highest level of aconitine (0.83%) was recorded in tubers collected from Phurkia Bugyal (3430 m) while the lowest values were found in samples from Kedarnath population (3600 m). The active principle content could not be correlated with the altitudinal difference.

Keywords: Aconitine, alkaloids, Himalayan alpines, medicinal herb, *Aconitum heterophyllum, A. balfourii*

Introduction

Aconitum heterophyllum Wall and *A. balfourii* Stapf (Ranunculaceae) are important medicinal herbs distributed in the alpine and sub alpine belts of the Himalayan region, at altitudes ranging from 2500 to 4300 m[1]. Their tuberous roots have been used by various ethnic communities for curing rheumatism, fever, neuralgia, etc. and are used in the preparation of different types of syrups in Indian systems of Ayurveda as well as in the Unani medicine[2]. The ever increasing demand in recent times for the naturals, has led to indiscriminate collection of medicinal plants from the wilds, and *A. balfourii* and *A. heterophyllum* are no exception; their status in the Himalayan region is 'critically endangered' as per IUCN category[3].

A number of alkaloids have been reported from various *Aconitum* species found in different parts of the world. Among the species of Indian Himalayan Region (IHR), isolation and/or structure elucidation have been carried out in *A. heterophyllum*[4], *A. balfourii*[5], *A. ferox*[6] and *A. falconerii* (syn: *A. tuberosum*)[7]. Qualitative and quantitative variations in the alkaloid profile of four *Aconitum* species, namely *A. heterophyllum, A. balfourii, A. violaceum*

and *A. falconerii* collected from Garhwal Himalaya have been reported[8]. HPLC analyses revealed that *A. falconerii* contains bisaconitine which is absent in other three Indian species. *A. balfourii* is the only species out of the four examined that contains pseudoaconitine as a major alkaloid, and *A. heterophyllum* and *A. violaceum* contain atisine. Since *A. violaceum* contains indaconitine in addition to atisine, these species can be differentiated from each other on this basis[8]. *A. balfourii* has been reported to be the main source of pseudoaconitine, balfourine, bikhaconitine and aconitine[5,8] while *A. heterophyllum* contains atisine, heteratisine and aconitine[4,8]. In view of the medicinal importance and 'critically endangered' status of these plants, it would be relevant to select elite plants based on active ingredient content, thus providing elite and quality propagules for commercial cultivation.

Material and Methods

Plant material

Daughter tubers of various sizes of *A. balfourii* (45-100 mm long and 10-30 mm diameter) and *A. heterophyllum* (10-30 mm long and 6-14 mm diameter) were collected in October (1998-2000) from various populations (Table 1) of Kumaun and Garhwal region of Indian Central Himalaya. The tubers were sprinkled with a systemic fungicide, Bavistin (50% carbendazim, w/v) and brought in

Table 1– Alkaloid content (% of dry wt.) in tubers of *A. heterophyllum* and *A. balfourii* collected from various populations of Garhwal and Kumaun Himalaya.

Population (Place)	Altitude (m)	*A. heterophyllum*	*A. balfourii*	
		Aconitine	Pseudoaconitine	Aconitine
Garhwal Himalaya				
Bharnala	3000	0.72	0.24	0.28
Dodital	3200	0.48	0.46	0.33
Goi	3200	NA	0.24	0.81
Dayara (Syari Bugyal)	3280	0.30	0.33	0.68
Hemkund	3300	NA	0.45	0.79
Tungnath	3600	0.14	NA	NA
Kedarnath	3600	0.69	0.39	0.13
Kumaun Himalaya				
Khatia	3250	0.67	NA	NA
Phurkia	3260	0.75	NA	NA
Kafni	3400	0.13	0.06	0.17
Phurkia Bugyal	3430	NA	0.62	0.83

NA= Not available at the time of collection; all values are average of 3 HPLC injections.

perforated polythene bags to the Institute. These were washed to remove traces of soil and dried at room temperature (22°C) for 20 days. The air dried tubers (at least 5 tubers of different sizes) were powdered in a grinder and made into a composite mixture before analysis.

Since pure standards of aconitine and pseudoaconitine were available, only these two compounds were estimated; *A. balfourii* was analysed for pseudoaconitine and aconitine while *A. heterophyllum* for aconitine.

Extraction

The samples (1.00 g dried powder) were extracted (25 ml x 3; 30 min each) with ammoniacal ether (ether containing 5% v/v, ammonia solution); the residue was then extracted with methanol (25 ml) for 16 h followed by two more extractions for 3 h each[9].

Column and thin layer chromatography

Samples were further purified on neutral alumina (Sisco Research Laboratories Pvt. Ltd., Mumbai) columns (8x2 cm; length & diameter) eluted with 50 ml of ethyl acetate and methanol (7:3, v/v)[9]. The eluates were dried *in vacuo* (30°C) in a rotatory film evaporator (Kinematica, Switzerland), dissolved in HPLC grade methanol (1.0 ml) for further analysis by HPLC, either directly or following further purification by thin layer chromatography (TLC). TLC was performed on 0.2 mm thick silica gel layer (Silica gel 60 GF_{254}; Merck, Germany) using cyclohexane, chloroform and diethyl amine (5:4:1, v/v)[10]. The portions of silica gel corresponding to the standard aconitine (Rf = 0.75) and pseudoaconitine (Rf = 0.66) were carefully scraped off and eluted (x3) with methanol (100%) containing 0.1% acetic acid (20 ml x 3). The elutes were dried *in vacuo* at 35 °C, and residue taken up in 0.3 ml of methanol for further analysis by HPLC.

High performance liquid chromatography

The quantification of aconitine and pseudoaconitine was carried out using a HPLC system (Kontron 322; Kontron Instrument Ltd., Italy) in RP-1- Spherisorb column (250 x 4.6 mm id, 5 µm; Merck Darmstadt, Germany), eluted in an isocratic mode with methanol and water (60:40, v/v) containing 0.1% of acetic acid. The column eluates were monitored using an online Kontron UV detector set at 263 nm. The peaks were identified on the basis of retention time and quantification was carried out on peak area basis using a dose - response curve prepared with authentic compounds. Three analyses were done per sample. The lower limit of detection was approx. 100 ng. Aconitine was obtained from Sigma Chemical Co., St. Louis, USA while pseudoaconitine was a kind gift of Prof. K.S. Khetwal (Department of Chemistry, Kumaun University, Nainital).

Results and Discussion

Analysis of tubers of *A. heterophyllum* and *A. balfourii* collected from different populations in Kumaun and Garhwal regions of Uttarakhand is summarized in Table 1. The levels of aconitine estimated in tubers of *A. heterophyllum* varied from 0.13-0.75% (dry weight basis) and the maximum value was detected in tubers collected from Phurkia (3260 m) while the lowest was detected in tubers from Kafni (3400 m). In *A. balfourii*, the levels of pseudoaconitine ranged from 0.06-0.62% (dry wt. basis). Highest level (0.62%) was recorded in tubers collected from Phurkia Bugyal (3430 m) while the lowest value (0.06%; about ten-fold less) was found in tubers from Kafni (3400 m). The amount of aconitine also varied and ranged from 0.13-0.83%. As in case of pseudoaconitine, highest aconitine levels (0.83%) were recorded in tubers collected from Phurkia Bugyal (3430 m).

Quantification of pseudoaconitine and/or aconitine in tubers of *A. heterophyllum* and *A. balfourii* indicated a wide variation amongst populations growing across a wide altitudinal range. However, the content of active ingredients could not be correlated with the altitudinal variation. Nevertheless, in one of the high altitude (> 3200 m) locations, i.e. Phurkia (in Kumaun Himalaya), the level of these active ingredients was maximum. In the present study, due to limited availability of propagules and in view of ensuring preservation of plants under natural habitat, only limited number of tubers of various sizes was randomly collected.

It is quite possible that the variation in alkaloid content could result from difference in the age of plants, as reported in *Taxus baccata* trees[11] and in *Podophyllum hexandrum* herbs[12,13]. The existing natural populations are likely to be of seed origin and thus the variation in active ingredient content could be due to genotypic differences. In contrast to this study, the amount of pseudoaconitine and aconitine content in *A. balfourii* and *A. heterophyllum* was reported to generally increase with altitude[8]. However, in the present study, the amount of pseudoaconitine and aconitine was higher than the values reported for the same species earlier[8]. Although ecological factors like habitat, temperature and soil characteristics would affect qualitative and quantitative changes in aconitine analogs[9,14], variations in the levels reported in different studies could also be attributed to the methods of purification and estimation used. A variation in the aconitine content of daughter tubers of *A. napellus* has also been reported to range between 0.3 to 2.0%[15,16]. It must be mentioned that the podophyllotoxin content in rhizomes of *P. hexandrum*, another medicinal herb, in different populations of Kumaun region indicated a positive correlation between podophyllotoxin content and an increase in altitude[12].

In conclusion, the results of this study highlight the need for mass scale multiplication of elite populations of species based on the active ingredient content for commercial cultivation. Further studies to explore populations with even higher content of active principle(s), and to examine if correlations exist in relation to habitat, soil characteristics, weather conditions and other factors vis-à-vis levels of these compounds would be important.

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Clastogenic assessment of two antibiotics on safflower

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Abstract

Nowadays due to wide spectrum effect of antibiotics in human beings, concern has been drawn towards its application in agriculture for the prevention and control of diseases without undergoing its adverse effects that it can cause if consumed in higher concentrations. Antibiotics are used in agricultural fields to boost up the yield but their high doses can cause genetic alterations in plants and can even make the plants sterile or incompatible.

In the present study, two antibiotics Streptomycin and Penicillin were found to induce various chromosomal aberrations like scattering, precocious movement, unorientation, bridge, micronuclei etc. in somatic cells of Safflower (*Carthamus tinctorius* L. var. PBNS 40) used as the test system at different concentrations. During the present investigation it was found that streptomycin was more potent antibiotic in causing chromosomal aberrations as compared to penicillin. Hence, there is a keen need to prevent excessive use of antibiotics otherwise the genetic integrity of the plant can be altered.

Keywords: *Carthamus tinctorius* L.var. PBNS 40, antibiotics, streptomycin, penicillin, chromosomal aberrations.

Introduction

The utilization of antibiotics in agriculture in place of pesticides is a latest trend to increase the yield and to provide better survivality to plants but it has negative effects too. Reports existed proves that the antibiotics can make the plants male sterile where the pollen grains are not functional, therefore the plants will set seed only when it is outcrossed. Streptomycin and Penicillin are commonly used antibiotics employed against bacterial diseases but their excessive doses may be harmful [1,2,3,4,5].

The present investigation has been carried out on Safflower, which is a unique plant because of its high linolenic acid content (>85%), not found in any other oilseed crops. Similarly Safflower oil naturally contains one of the lowest levels of total saturated fatty acids (palmitic acid and stearic acid) amongst oilseed species[6]. Saturated fatty acids are undesirable in edible oils because of their hypercholesterolemic effect[7] since the safflower oil has the capacity to reduce coronary heart disease[8] and it lowers cholesterol level, so excessive use of antibiotic i.e. beyond tolerance limit on plants brings chromotoxicity which results in alterations in oil characteristics and thus prove to be toxic to humans also.

Therefore keeping in view all these facts, attention should be drawn towards the sensitivity of plants against these antibiotics. So, the present study is an attempt to assess the mitotoxic effect of antibiotics on somatic cells of Safflower.

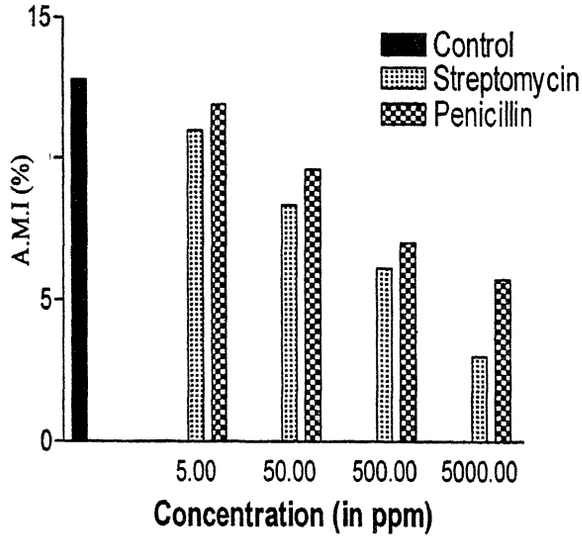
Material and Methods

Solutions of streptomycin and penicillin were prepared for four concentrations 5, 50, 500, 5000ppm for each antibiotic. For the mitotic study healthy inbred seeds of Safflower were presoaked in distilled water and then allowed to germinate. The germinated seeds were then pretreated in respective concentrations of antibiotics for 3 hours. After the treatment, they are washed thoroughly and fixed in Carnoy's fixative (1:3 glacial acetic acid: acid

alcohol for 24 hours and then transferred in 70% alcohol. Untreated root tips are fixed and used as control. Slides were prepared using chromosome squash technique with 2% acetocarmine and photo-micrographs were analyzed using computerized Nikon Image capturing system.

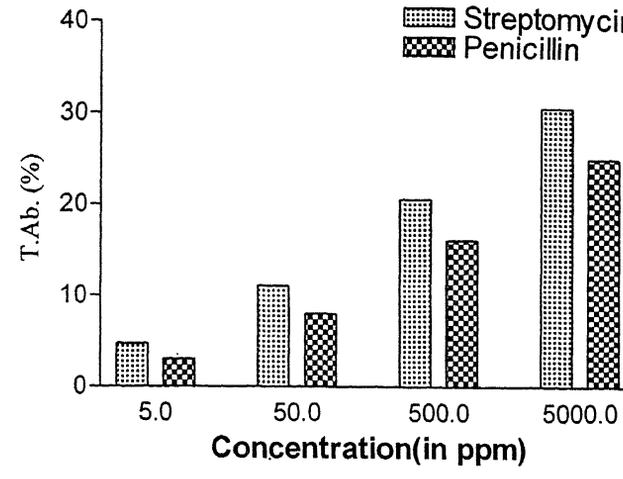
Results

The effect of each antibiotic on the mitotic index has been analyzed separately in Table-1. The table shows that the mitotic index was 12.80% in the control set with no reports of chromosomal aberrations (Fig a, b). But, as the concentration of antibiotics increased and the time prolonged, the Active Mitotic Index (AMI) showed gradual decrease. At various concentrations of treatments viz. 5, 50, 500, 5000ppm the decrease in AMI ranged from (11.00%-3.02%) in case of streptomycin and (11.92%-5.83%) in case of penicillin treated sets as shown in (Graph 1). Antibiotics not only inhibited the rate of mitosis but also in most cases percentages of abnormal mitotic phases were found to increase alongwith the increasing concentrations.



Graph 1–Showing comparative effect of Streptomycin and Penicillin on Average Mitotic Index (A.M.I.)

Chromosomal aberrations at metaphase viz. stickiness, precocious movement, anaphasic bridges and micronuclei etc. were more common nearly all the treatment sets but their frequencies were dose dependent. The type of chromosomal aberrations observed in treated sets included unorientation (Fig-c), scattering (Fig-d), precocious movement (Fig-e), stickiness (Fig-f), double bridge with unequal separation (Fig-g), micronuclei (Fig-h), and trinucleate cells (Fig-i) etc.



Graph 2–Showing comparative effect of Streptomycin on Total Abnormality percent (T.Ab%)

As regards the mitotic frequency it was found that streptomycin had affected the mitotic spindle and increased metaphasic stages as compared to anaphasic and telophasic stages. However in case on penicillin treated sets chromosomal abnormalities were less vigorous. Maximum chromotoxicity was recorded to be 30.80% in case of streptomycin treated sets and 24.89% in case of penicillin treated sets at 5000ppm dose (Table-1).

Discussion

The cytogenetic effect of different antibiotic treatments on mitotic division of root tip cells of Safflower has been presented in Table-1. The present analysis reveals that higher concentration of both the antibiotics i.e. streptomycin and penicillin is harmful to plants but their degree of toxicity individually has been differed which helped us to conclude that streptomycin is more chromotoxic as compared to penicillin as shown in (Graph 2) so their use should be restricted within tolerance limits

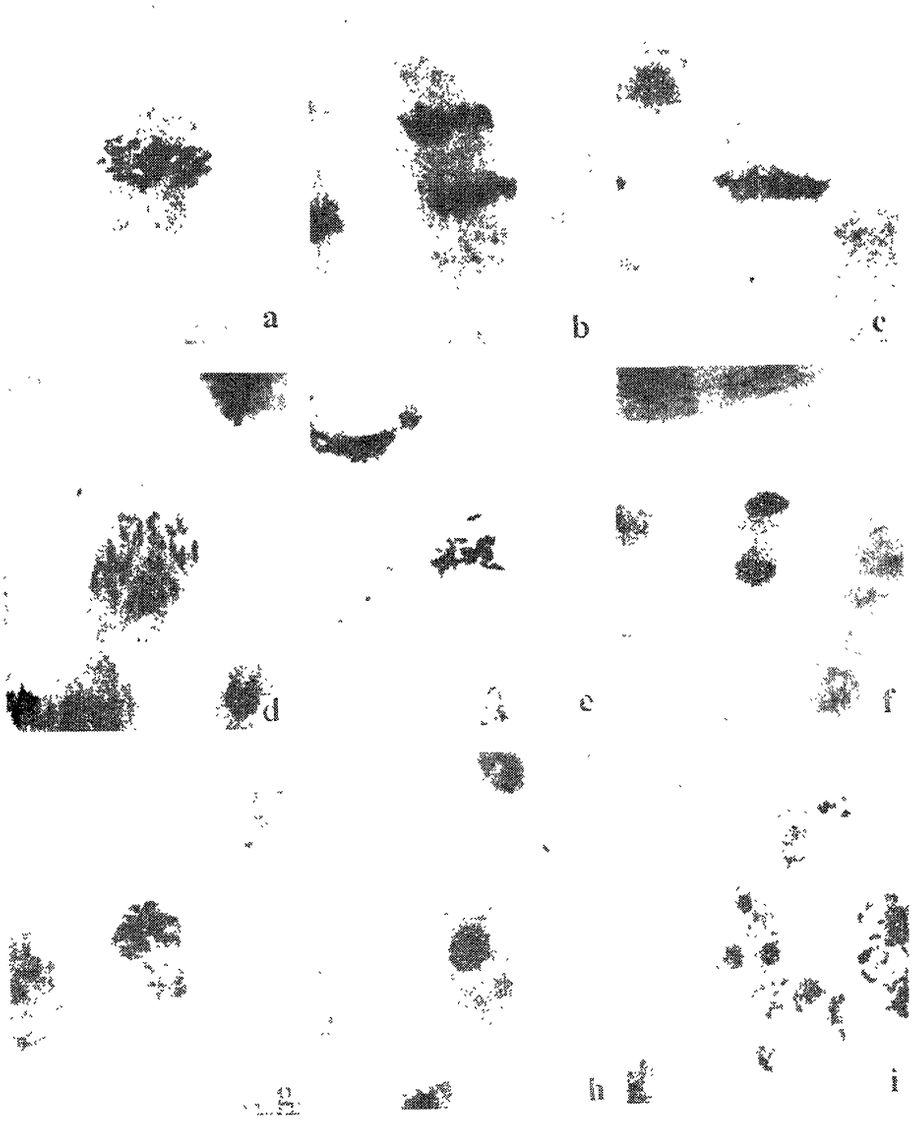


Fig a– Normal Metaphase (2n=24); Fig b– Normal Anaphase (24:24 separation); Fig c– Unorientation at Metaphase; Fig d– Scattering at Metaphase; Fig e– Precocious movement at Metaphase; Fig f–Stickiness at Anaphase; Fig g– Double Bridge at anaphase with unequal separation; Fig h–Micronucleus; Fig i– Trinucleate cell

i.e. in small proportions. On increasing the concentration mitotic indices shows a gradual decrease and an increase in abnormality percent, which suggests that the antibiotics interfered with normal sequences of mitosis. Such a reduction in mitotic activity may be due to inhibition of DNA synthesis [9] or it may be due to gene mutation[10]. Stickiness has been attributed to improper folding of chromosome at any phase of cell cycle, which makes the chromatids connected by subchromatid bridges[11]. Precocious movement arises due to movement of chromosomes ahead of rest[12] or due to early terminalization of chiasma. Bridges at anaphase may be suspected due to stickiness at

Table 1– Various Chromosomal Abnormalities at Different Doses of Antibiotic Treatment In *Carthamus tinctorius* L. var. PBNS 40.

Treatment	Conc.	Metaphasic Abn. (%)					Anaphasic Abn. (%)					T.Ab	A.M.I
	ppm.	Un	Pm	St	Sc	Oth	Br	St	Mn	Us	Oth	(%)	(%)
Control	-	-	-	-	-	-	-	-	-	-	-	-	12.80
Streptomycin	5	0.25	0.93	0.72	0.63	0.58	-	0.42	0.51	-	0.68	4.72	11.00
	50	0.47	1.68	1.02	1.47	1.88	0.92	1.00	1.02	0.36	1.23	11.05	8.33
	500	1.62	2.58	2.93	1.89	2.36	2.46	2.11	1.82	0.68	2.00	20.47	6.12
	5000	2.12	3.47	3.78	2.92	3.01	3.81	3.96	2.71	1.36	3.81	30.38	3.02
Penicillin	5	0.20	0.81	0.62	0.31	0.25	-	0.30	0.20	-	0.31	3.00	11.92
	50	0.59	1.43	1.02	1.36	0.85	0.21	0.82	0.68	0.41	0.64	8.02	9.62
	500	1.92	2.62	1.92	2.95	1.18	1.11	2.44	1.85	0.93	1.00	16.00	7.01
	5000	2.01	3.48	3.55	3.00	2.00	2.17	3.54	2.36	1.12	1.66	24.89	5.73

Abbreviations: Conc. Concentration, Abn. Abnormality, Un. Unorientation, Pm. Precocious movement, St. Stickiness, Sc. Scattering, Oth. Other, Br. Bridge, Mn. Micronuclei, Us. Unequal separation, T.Ab. Total abnormality, A.M.I. Average Mitotic Index.

metaphase and their failure to separate at anaphase or due to breakage and reunion of chromosome[13,14] Clastogenic type of abnormalities like micronuclei and trinucleate cells may be used as parameters to trace out mutation activity[15]. Micronuclei observed may have arisen through laggard chromosome. So, the present paper reports that caution should be done to conserve the efficacy of antibiotics in human medicines that scrutiny to all antibiotic uses, including uses on plants.

Our study presents noteworthy results regarding efficacy of these two antibiotics which cannot be neglected because any damage to plants may be transferred to human beings and proves to be fatal hence lower doses of antibiotics should be recommended for agricultural use.

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Application of diffusion technology for seed identification, determining critical time for germination, water diffusivity and enhancing germination of maize(*Zea mays. L*) genotypes

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Abstract

Diffusion technology developed earlier on the basis of the theory of diffusion concerning electrolyte/non-electrolyte (1987) has been further employed for determining seed constant for seed identification of two Ethiopian maize (*Zea mays.L*) genotypes *viz.* Rare -1 and Melkassa – 1 in our laboratory. The seed constant values obtained for the above genotypes are $0.1373*10^{-3}$ and $0.0847*10^{-3}$ respectively indicating clearly that the two maize varieties are genetically different from one another. The new theory of water diffusivity (2007) recently developed in the laboratory has also been applied for the determination of the critical time for germination and water diffusivity and these are found to be 15h, 141h, $0.2206*10^{-5}$ cm^2s^{-1} and $0.2063*10^{-5}$ cm^2s^{-1} respectively for the genotypes at 25^0C. Laboratory germination test using bulk seeds and the above critical time for germination showed 100% germination for the genotypes and there was an increase of 6% and 8% in comparison to control experiments carried out under similar experimental conditions. These studies clearly showed that the critical time for germination determined by using the diffusion technology has improved the germination of the two maize varieties. The results are compared and discussed with those obtained earlier for other maize and wheat genotypes. The importance of diffusion technology for further work and applications in the area of seed science and technology are also highlighted.

Keywords: water diffusivity, maize genotypes

Diffusion concerning water uptake in germinating seed and plant is of fundamental importance for the growth, development and crop production. Earlier models on water absorption are either based on the relative concept of water potential or water content and mostly assumed the seed to be of spherical geometry[1-6]. Some years earlier a serious controversy has arisen on the use of the water potential concept and which has been found to be problematic[7-12]. In addition to this the accuracy obtained in the determination of water diffusivity of germination seed was confined to the faulty assumption of assuming the seed to be a sphere and therefore the data obtained are in errors[1-6]. Also most of the experimental values are determined under non-steady state conditions and are therefore not comparable. Further all seed germination experiments determine only the average time for germination and provide no information about the critical time for germination and critical moisture attained during this time although these are of great significance for our understanding of the germination process. Therefore, a proper co-relation needs to be established between the critical time for germination, critical moisture content, percent germination and crop production. Finally no work has been undertaken for using diffusion process for seed identification although it is clear that different genotypes of a crop are different in composition, structure and also physiologically. With these objectives in mind a theory of diffusion concerning electrolyte/non-electrolyte for germinating seed was earlier developed[13] and methodology also established[14-16] for seed identification on the basis of seed constant. However, the above theory is not applicable to water diffusion (uptake) studies because of the fact that the driving force for diffusion in Fick's law has to be restated as the molar concentration of a given amount of water remains almost constant. Therefore, a new theory of water diffusivity was recently formulated[17] which is based on particle density gradient concept rather than concentration gradient. The validity of the theory was established by carrying out experiments using soybean, maize and wheat genotypes and also the concept of critical time for germination and critical moisture content developed and established on the basis of steady-state conditions. Water diffusivity, critical time for germination and critical moisture content were further co-related with

percent germination and enhancement in germination established for some genotypes of the above crops[17-19].

In this article work has been further extended by carrying out water uptake studies at 25°C for two maize genotypes *viz.* Rare-1 and Melkassa-1 and co-relation established between seed constant, water diffusivity, critical time for germination and percent germination. The results are discussed and compared with those obtained for other maize and wheat genotypes[17-19] and the application of the diffusion technology for further work in the area of seed science & technology highlighted.

The theory of water diffusivity[17] in which Fick's law has been restated in terms of particle density gradient in place of concentration gradient is mathematically related to the flux and diffusion constant *(D)* as

$$J(t) = -D\left[\frac{\partial N}{N_A}\right] / \partial X \qquad (1)$$

where N is the number of water molecules per unit volume and N_A is the Avogadro's constant. For diffusion in germinating seed the theory developed relates the average integral diffusion coefficient $\overline{D}$ by the equation

$$\overline{D} = \frac{1}{\beta_{seed} t} \ln \frac{\Delta N_0}{\Delta N_f} \qquad (2)$$

where β_{seed} is the seed constant defined by

$$\beta_{seed} = \frac{A}{l}\left(\frac{1}{V_1} + \frac{1}{V_2}\right) \qquad (3)$$

in which A and l are the effective area and pore length of the seed membrane/coat and V_1 and V_2 are the volumes of the water and that of the seed respectively. $\Delta N_0 = N_1 - N_2$ and $\Delta N_f = N_3 - N_4$ are the initial and final number of water molecules before and after the diffusion process and t is the time of

Table1– Seed constant (β_{seed}), water diffusivity ($\overline{D}$) and percent germination of Rare-1, Melkassa-1 and for some other maize, wheat and soybean genotypes at 25°C

Crop	Genotypes	Seed constant (β_{seed} x 10^3)	Water diffusivity ($\overline{D}$ cm^2s^{-1}) x10^5	Critical time for germination(t_{ctg})h	Percent germination	
					Experiment	control
Maize	Rare 1	0.14	0.2206	15	100	94
	Melkassa-1	0.08	0.2063	141	100	92
	DPT-1	6.8	1.3478	70	80	76
	Al-Comp	0.38	0.3191	53	66	50
	BH-660	105.0	0.0651	136	87*	79*
	Pop 902x903	132.2	0.0588	63	68*	66*
	Paras**	233.3	0.0278	95	95	80
	Parkash	175.6	0.0323	93	95	80
Wheat	Har-1781	8.2	0.2736	49	98	92
	Har-3354	53.7	0.3800	41	89	74
	Simba	2.7	1.2379	45	98	88
Soybean	PK-416**	276.8	0.0339	74	95	80
	SL-295**	287.6	0.0365	69	90	85

*Field trials others are laboratory germination test
**at 30°C

diffusion in seconds. Therefore to determine $\overline{D}$, β_{seed}, t, ΔN_0 and ΔN_f must be known.

Determination of seed constant (β_{seed})

Eqn. (3) defines the seed constant with A, l. V_1 and V_2. However the seed constant cannot be directly estimated by this equation as it is not possible to determine A and l precisely. Therefore an alternative procedure has to be used. In our earlier studies[14] seed constant was determined by diffusing a reference electrolyte, 0.01 M KCI into seed for which the exact value of $\overline{D}$ in water is known. This was necessary since the exact $\overline{D}$ for water in a seed is not known.

The procedure to determine the seed constant and the experimental apparatus and the theory of diffusion developed for electrolytes/ non-electrolytes are given elsewhere[13-14]. It is important to note that seed constant in a seed can be determined by employing any suitable concentration of KCI and not only 0.01 M KCI as quoted in our earlier paper but we retain this concentration as one of the best options for all seed constant determination for uniformity. In the present work seed constant of Rare-1 and Melkassa-1 *(Zea mays. L)* genotypes were determined by using 0.01 M KCI at 25^{0}C. The values of β_{seed} are recorded in Table 1.

Procedure : *Water Diffusion Experiment with Maize (Zea mays. L) Genotypes*

(i) Diffusion Cell

The diffusion cell designed[13] earlier and later modified[14] was employed in the present study.

(ii) Diffusion runs with water – determination of $\overline{D}$ *at 25*°C

Maize seeds of Rare-1 and Melkassa-1 (*Zea mays. L*) genotypes having fixed initial moisture contents were procured from the Department of Plant Science, Haramaya University, Ethiopia and were preserved in air tight containers at room temperature. From these lots seeds having nearly constant weights (up to third place of decimal) for each genotype were carefully selected and a single grain was used for each water diffusion experiment at 25 ± 0.5^{0}C. The diffusion cell was thoroughly cleaned and dried. A single seed was then carefully inserted into the bottom of the cell with the help of a pair of clean tweezers so that no injury is caused to it. The cell was next filled with pure water having a conductance of ~49μS and which was thermo-stated before hand. The B-19 standard joint of the diffusion cell and to which high quality silicon grease was slightly applied was slowly and carefully inserted so that no air bubbles remained in the cell. The cell was then thermo stated in an upright position maintained at 25 ± 0.5^{0}C and the diffusion run timed from this point. After the attainment of steady-state conditions the experiment was stopped and time for diffusion recorded. The monitoring of steady-state conditions was on the basis of seed exudation process the details for which are discussed elsewhere[17]. After the diffusion experiment the cell was removed from the thermostat and the seed taken out carefully with the help of a pair of clean tweezers. Utmost care was taken to remove the B-19 male joint from the cell so that no water droplets remained sticking on it. Also the seed with the tweezers was carefully handled and this was given few slight jerks over the mouth of the diffusion cell so that every excess droplet of water sticking on its surface falls back into it. The seed was immediately weighed and once the final weight of the seed in the three experiments was taken eqn. (2) was used to determine $\overline{D}$ since β_{seed}, time, t in seconds, initial and final weights of water (g) from which N_1, N_2, N_3 and N_4, the initial and final number of water molecules can be easily determined by the use of simple relation:

$$\text{Number of water molecules} = N_A * n$$

where N_A is the Avogadro's number and n is the chemical amount of water. In Table 1 are also recorded the $\overline{D}$ values obtained. The $\overline{D}$ values recorded for the maize genotypes are the mean of three replications at 25°C.

(i) Maize genotype identification

The earlier theory[13] of diffusion concerning electrolyte/non-electrolytes of germinating seed involved an important constant called seed constant and symbolized as β_{seed} and which is a specific characteristic of seed coat/membrane of a genotype. The methodology for seed identification has been

established earlier in the laboratory and seed constant were determined for a number of maize, wheat and soybean genotypes[13-19] In Table 1 are recorded the seed constant obtained earlier for some maize, wheat and soybean genotypes along with that of Rare-1 and Melkassa-1 maize genotypes undertaken in the present study[13-19]. From the results it is clear that the seed constant for Rare-1 and Melkassa-1 are quite different from those obtained for other maize genotypes indicating clearly that these are genetically different and hence are the fingerprint of the above two genotypes. Seed constant is now considered to be a more complex constant and its determination from diffusion experiment is not only related to seed characteristics but also has its physiological contribution as well. In our earlier studies[13-19] seed constants of genotypes were determined by employing single seed of same age, moisture content, weight and from the same lot. But all seeds of the lot cannot be of same size and weight and therefore obtaining only one constant value for the seed constant was a problem and the water diffusivity obtained also will not be the true constant for all the seeds. To overcome this difficulty it is now considered important to determine seed constant for a number of seeds of a genotype which are of different weight and size but are of same age, moisture content and from one lot and then to obtain the statistically average value. This is important as the time for attaining steady-state conditions for different seeds of some genotypes will lie within a certain range and need to be averaged for uniformity. This process of seed identification using diffusion technology will only then become universal in nature and can be fruitfully exploited for seed fingerprinting of any crop genotype.

(ii) Water diffusivity

The water diffusivity ($\overline{D}$) obtained for the two maize variety *viz.* Rare-1 and Melkassa-1 are recorded in Table 1 and these are $0.2206 \times 10^{-5} cm^2 s^{-1}$ and $0.2063 \times 10^{-5} cm^2 s^{-1}$ respectively at 25^0C. In Table 1 are also recorded the $\overline{D}$ values for some other maize genotypes as well as those of wheat and soybean crops obtained earlier in the laboratory[13-19]. A glance at the table indicate that the water diffusivity of the above two maize varieties are of similar orders of magnitude not only with those of other maize genotypes but also with the wheat and soybean genotypes indicating clearly the validity of the theory of water diffusivity based on particle density gradient concept. Phillips[2] using T204 x C121 corn and employing the theory and procedure developed obtained an average value of water diffusivity of 7.91×10^{-5} $cm^2 h^{-1}$ (which is 0.022×10^{-5} $cm^2 s^{-1}$) after 36h water uptake at 28^0C without attaining steady-state conditions and which is also of the same orders of magnitude as obtained for Rare-1 and Melkassa-1 taken in the present study. Similar order of magnitude was also obtained earlier for maize by Chittenden and Hustralid[4] *viz.* 0.001×10^{-5} cm^2 s^{-1}. A comparison of the water diffusivity obtained by Phillips[2] and Chittenden and Hustralid[4] indicate a difference of $0.021 \times 10^{-5} cm^2 s^{-1}$ which is 22 times higher whereas with those obtained for Rare-1 and Melkassa-1these differences are $0.1886 \times 10^{-5} cm^2 s^{-1}$ and $0.1865 \times 10^{-5} cm^2 s^{-1}$ resulting in ten and nine times higher values. These differences might have arises due to the fact that:

(i) no steady-state conditions were obtained by Phillips[2] due to lack of proper experimental design whereas in the present work a diffusion cell was employed and steady-state conditions attained.

(ii) the model of Phillips[2] was based on the faulty assumption of assuming seed to be of spherical geometry due to which errors in diffusivity values may occur because of the departure of the geometry of seeds from perfect sphere. But the recently formulated theory[17]of water diffusivity assumed seed to be any shape and size and therefore the chances of errors are minimized.

(iii) The methodology used by Phillips[2] in the experiments involve periodic wiping of seed surface using cotton towel which may result in non-uniform preferential absorption of surface water from the pores of the seed coat/ membrane due to capillary action of the cotton towel employed and there are every chances of obtaining erroneous water diffusivity. To verify this aspect experiment was also carried out in one of our earlier study[19] with BH-660 and Pop 203x203 maize genotypes the seeds of which were wiped only once after its removal from the diffusion cell on the attainment of steady-state conditions.

The results obtained have shown that the $\overline{D}$ were found to be lower by $0.0045x10^{-5}cm^2s^{-1}$ and $0.0064x10^{-5}cm^2s^{-1}$ indicating that even with only single wiping of the seed surface the amount of water removed from the surface is significant and in case of Phillips[2] experiments since periodic wiping were done more errors might have introduced resulting in erroneous water diffusivity. This also make it clear that the methodology used in the present study in which no wiping was necessary except at the end of the experiment is more reliable and chances of introducing any serious error is therefore negligible.

From our present experiment the amount of water absorbed was also quantified before and after attainment of steady-state conditions and it was found that Rare-1 absorbed nearly 1.2 times more water as compared to Melkassa-1 i.e. 0.0829g and 0.06910g respectively at 25^0C and the $\overline{D}$ value is also 1.07 times higher which explains the reason of higher water uptake by it.

Critical time for germination (t_{ctg}) and its application for enhancing germination.

As discussed earlier[17-19] a new term *viz.* critical time for germination symbolized as t_{ctg} was obtained in water diffusion experiment and which can be easily extended to any solution. Accordingly this critical time for germination is very specific for a particular seed genotype since it is at this time the seed is assumed to attained a certain fixed moisture level which may now be called 'critical moisture level or content' and can be symbolized as 'C_{mc}'. It is at this stage that the seed is at the peak of its physiological and biochemical activities necessary for germination. It is presumed that once the seed attained the above critical time for germination, the seed if tested for germination should germinate provided minimum moisture level exist in the soil so that the seed after attaining the C_{mc} should not dehydrate otherwise there is a possibility of germination getting effected. In order to check the above hypothesis work carried out earlier with other maize, wheat and soybean genotypes[17-19] (cf. Table 1) have clearly shown that the critical time for germination really enhanced germination in all cases. In all the experiments bulk seeds of genotypes were first made to hydrate up to the critical time for germination determined in the diffusion experiments and then immediately sown in soil maintained prior with sufficient moisture content in the laboratory germination tests. In the present work identical procedure was adopted and the critical time for germination *viz.* 15h and 141h was experimentally determined with two maize varieties. Laboratory germination tests with bulk seeds and in three replications were next carried out at room temperature (which varied from 15^0 to 26^0C in 24h) and the percent germination determined which came out to be 100% in comparison to control experiments carried out with un-soaked (no diffusion) seeds under same identical conditions. The control germination tests gave 94% and 92% for Rare-1 and Melkassa -1 respectively showing clearly that there was enhancement of germination by 6% and 8% for the two genotypes. The critical time for germination for other maize wheat and soybean genotypes obtained earlier in the laboratory are also recorded in Table 1.

Conclusion

From the present study using two maize genotypes *vi.z* Rare-1 and Melkassa-1 it can be safely concluded that the validity of the theory of water diffusivity recently formulated in the laboratory is further established. The seed constant obtained for the two genotypes also clearly showed that these two genotypes are genetically different and they absorbed water at different rates. It has been found that Rare-1 maize genotypes absorbed nearly 1.2 times more water in comparison to Melkassa-1 at 25^0C.

Further it is now possible to quantify the critical moisture content symbolized as C_{mc} from the critical time for germination (t_{ctg}) which can be obtained from diffusion experiments for any seed of a genotype and of any crop. It is also clear that the average time for germination (t_g) obtained by Phillips for maize and other seed genotypes is in no way equal to critical time for germination (t_{ctg})which is important for a seed to germinate. The importance and significance of t_{ctg} for the two maize genotypes for enhancing germination has again been clearly demonstrated for the two maize genotypes taken in the present study and can be fruitfully exploited for all work related to seed germination.

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The theory of the quantum Hall effect

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Abstract

Laughlin's theory of fractional charges is worked out in detail for small charges from 1/3 till 1/101. There is a small deviation between computed values and those obtained from the closed form expression. The ground state energy crosses that of the charge-density waves. We develop a theory of fractional charges by using the quantum mechanics of angular momentum. We find that fractional charges can be expressed in terms of spin and the values of charges 0, 1, 1/3, 2/3, 2/5, 3/5, ..., are produced. The angular momenta eigen values when subjected to flux quantization, yield plateaus of energies which are independent of the magnetic field. In this way we are able to predict that charges of ±2e, ±6e, ±10e, ±14e, ..., are produced. The higher order term in the flux quantization also produces a quasiparticle charge of ±4e. These calculated values of the charges are the same as those found in the experimental data of quantum Hall effect in graphene, which is a mono-atomic layer of carbon. Since the charge of the quasiparticles appears in the resistivity and there is a strong need of the electron spin to predict these charges, spin-charge coupling occurs in a natural way. We have examined the Dirac equation for this type of charge problem in which not only positively and negatively charged particles, electrons and positrons, but zero and fractional charges also emerge. It is found that the usual Dirac matrices become 8x8 instead of the usual 4x4 matrices. In this way, quasiparticles of many different charges are found.

Keywords: quantum Hall effect, fractional charges, graphene

Introduction

In 1983, Laughlin[1] wrote down the wave functions of the form $(z_i-z_j)^m$ with $z=x+iy$ and from that showed that the accumulated charge at a point is $1/m$ so that for $m=3$, the quasiparticle charge becomes $(1/3)m$. Laughlin also calculated the ground state energy which for $m=3$ turned out to be more favourable than that of the charge-density waves. The state described by ψ_m is incompressible because compressing or expanding it is tantamount to injecting particles. The Hall conductance is e^*e/h so that the effective charge of a quasiparticle is $e^*=e/m$ which for $m=3$ is $e/3$. The flux quantization is used to quantize the resistivity. In 1980, von Klitzing[2] found that the Hall conductivity shows a plateau. The plateau in the resistivity gives the correct value of h/e^2. Tsui *et al.*[3] discovered that plateaus in the Hall reistivity also occur at $h/(1/3)e^2$. Hence, there is clearly a need to find a theory of fractional charges. In the Stormer's data[4], the fractional charges found are, 1/3, 2/3, ... Recently, the Hall effect measurements have been performed with mono-atomic layer of carbon atoms called graphene. This material gives very large charges, $2e$, $4e$, $6e$, $10e$, $14e$, ... Therefore, it is important to understand fractional as well as large integral multiples of charge. We have found [5-14] that spin leads to factors which couple the charge so that the electron charge is seen to split. By the same theory it is possible to obtain large charges such as $2e$, $4e$, $6e$, $10e$, ...

In the present communication, we develop a theory to understand fractional as well as large charges. We use the quantum theory of angular momentum to derive the effective charges. We find that there is a strong spin-charge coupling so that the fractional charges found in the experimental data of quantum Hall effect originate from the spin.

Laughlin's theory

We describe the Laughlin's theory of fractional charges and extend the calculations from $m=3$ to 101. The electrons are confined to the x-y plane and

the magnetic field is applied in the z direction. The states of the lowest Landau levels are given by,

$$|m>=(2^{m+1}\pi m!)^{-1/2}z^m\exp(-\frac{1}{4}|z|^2) \quad (1.1)$$

where $z=x+iy$. The many-body Hamiltonian is,

$$\mathcal{H}=\Sigma_j\{|(\hbar/i)\nabla_j-(e/c)A_j|^2+V(z_j)\}+\Sigma_{j>k}e^2/|z_j-z_k| \quad (1.2)$$

where j and k run over N particles and $V(z_j)$ is the potential generated by a uniform neutralizing background. The ground state is written as,

$$\psi=\{\Pi_{j<k}f(z_j-z_k)\}\exp(-\frac{1}{4}\Sigma_l|z_l|^2) \quad (1.3)$$

which is antisymmetric for $f(z)=z^m$ with m = odd number. The total energy per particle in terms of a radial distribution function $g(r)$ in a plane is given by,

$$U_{tot}=\pi\int(e^2/r)[g(r)-1]rdr. \quad (1.4)$$

with $g(r)=1-\exp[-(r/R)^2]$. The ground state energies for $m=3$ and $m=5$ are slightly deeper than for charge-density waves and the integrated value is found to be,

$$U_{tot}=\frac{0.814}{\sqrt{m}}(\frac{0.230}{m^{0.64}}-1)\frac{e^2}{a_o} \quad (1.5)$$

The integrated values of the total energy from (1.4) is indeed very close to that of the formula (1.5) given by Laughlin. Write, $|\psi^{+z_o}|^2$ as $\exp(-\beta\phi')$ with $\beta=1/m$ and,

$$\phi'=\phi-2\Sigma_l ln|z_l-z_o| \quad (1.6)$$

which describes an object of which the charge is centered at z_o. The charge accumulated at z_o is $1/m$. The flux quantization is confined to the area, A, so that the quantized field is determined by,

$$B.A=\frac{hc}{e} \quad (1.7)$$

The system of flux-quantized electrons in the area A generates Hall currents without compressing and then a critical stress collapses the system by the quantum area, $m2\pi a_o^2$.

The length from (1.7) is $A^{1/2}=(\frac{hc}{eB})^{1/2}$ so that the collapsed area becomes,

$$A_{collapse}=m\ 2\ \pi\frac{hc}{eB}=\frac{2\pi hc}{(e/m)B} \quad (1.8)$$

which means that charge is replaced by e/m. For $m=3$, a particle of charge $e/3$ nucleates. Unfortunately, there is a catch. According to (1.7) only the product of the area and charge is a constant,

$$Ae=hc/B \quad (1.9)$$

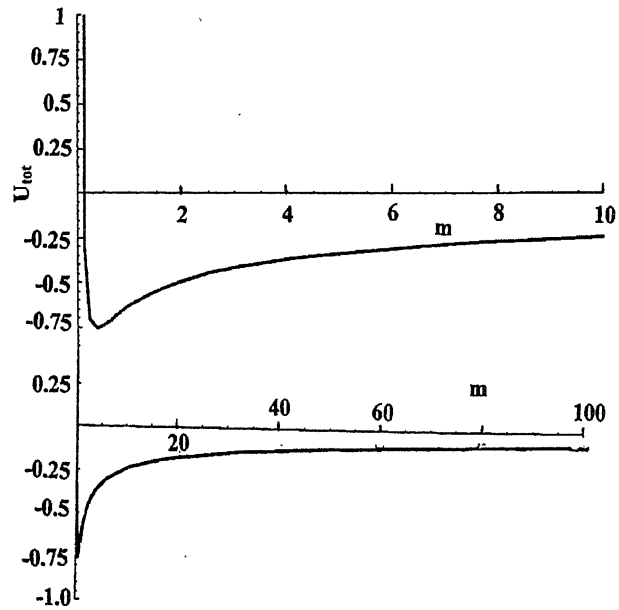

Fig. 1– The total energy of the Laughlin's solution upto the charge of 1/101 in units of e^2/a_o.

If area is multiplied by 3 and charge is divided by 3, then the equation remains unchanged. The unit flux hc/e also can be multiplied by an integer so that hc/e is to be replaced by nhc/e (n=integer). Therefore, charged particles can not be nucleated without the stringent requirement of incompressibility. We examined the ground state energy by using (1.5) as well as by numerical integration of (1.4) with the use of radial distribution function given by Jancovici[16] for m=0 to 101. From this calculation we learn that the ground state energy as a function of filling factor 1/m is a smooth function and does not have peaks except at m=0 which is a divergence. At m=3 (ν=1/3), it varies smoothly. Laughlin emphasizes that the energy values for m=3 are deeper than those of charge-density waves and at m=5 also, apparently the value may be deeper than for CDW. From (1.4) for m=5 the ground state has the energy of -0.3340 e^2/a_0 whereas for charge density waves the calculated value is -0.322 e^2/a_0. The difference between the two values is $0.018e^2/a_0$ which is barely 6 per cent. Usually, the energy as a function of a parameter should show minima at least at m=3 but the graph of U_{tot} from (1.4) does not have a minimum at m=3. Similarly, at m=5, U_{tot} does not have a minimum. The formula (1.5) is very close to numerically correct integrated value. We show the total energy as a function of m in Fig.1. The energy is positive for very small values of m becoming negative, going through a minimum and then increasing to a saturation value. The value goes through m=3 very smoothly. We find that the difference between (1.4) and (1.5) is within 5 %. Laughlin's is therefore a theory by which quasiparticles of fractional charges may be nucleated. This theory seems to be similar to that of α decay in which particles are generated by clustering in the nucleus.

The series of charges

Stormer-1: The fractional charges, 1/3, 2/3, 2/5, 3/5, 3/7, 4/7, ..., have been reported. The series of fractions 1/3, 2/5, 3/7, 4/9,4/7, 3/5, 2/3, 2x2/5, 2x2/3, are clearly seen in Fig. 18 p.886 of Stormer[4] revealed in 1999. The data at earlier times showed the emergence of fractional charges of 1/3 or 1, 2, ..., but the full series was identified by Stormer in 1999 as shown in Fig. 2. Stormer claims that one series observed is, $\nu=\frac{p}{2p+1}$, and the other series is $\nu=\frac{p}{2p-1}$ irrespective of the physics of the problem. In 1985, we tried to see if the charge can become fractional. There are electrons and there is a magnetic field so what can happen. In a simple way, we write the Bohr magneton $e\hbar/2mc$ so that any factors which multiply the Bohr magneton can influence the charge.

Stormer-2: The charges 2, 6, 10, 14, 18, ..., have also been reported. The quantum Hall effect measurements in a device made of graphene which is two dimensional mono-atomic sheet of carbon gives plateaus at ±2, ±6, ±10, ±14, ..., etc. When the magnetic field is increased to 25 Tesla, the series changed to ±2, ±4, ±6, ±10, ... The plateaus at 0, ±1 are also present[17,18] as seen in Fig. 3. Therefore, to understand this type of Hall effect is a nontrivial problem. We will see that simple use of quantum mechanics gives the correct series. Laughlin's theory does not deal with large charges such as 2*e*, 6*e*, 10*e*, 14*e*, 18*e*, ..., etc.

Theory of quantum Hall effect

Usually the free electrons are in the $l=0$ state so that the electron gas models give a single value of the charge of the electron. In these experiments $l=0$ is the root cause of observing a single value for the electron charge. We consider (a) the finite value of l and more spin symmetries than are usually taken into account. We allow two levels for s=1/2 and two more levels for spin = −1/2 so that there are four levels for spin ½. These levels are not superimposed on each other because there are two separate g values, one for spin +1/2 and the other for spin -1/2. We consider the spin as well as the orbital motion so that,

$$g_j j=g_s s+g_l l=\frac{1}{2}(g_l+g_s)j+\frac{1}{2}(g_l-g_s)(l\text{-}s). \qquad (2.1)$$

Multiplying both sides of the above equation by $j=l+s$ and taking eigen values, we find,

$$g_J j(j+1)=\frac{1}{2}(g_l+g_s)j(j+1)+$$
$$\frac{1}{2}(g_l-g_s)[l(l+1)-s(s+1)]. \quad (2.2)$$

Which upon substituting s=1/2 gives j=*l* ± *(1/2)* for which,

$$g_J = g_l \pm \frac{g_s - g_l}{2l+1}. \quad (2.3)$$

For g_s=2, g_l=1 we find,

$$g_\pm = 1 \pm \frac{1}{2l+1} \quad (2.4)$$

Usually the treatment of cyclotron resonance is for free electrons but we need to introduce the concept of *g* values in the cyclotron resonance also. The usual expression is, $\omega = eB/mc$. Corresponding to this frequency, the voltage along y direction is $\hbar\omega = eV_y$. From the last two expressions, $\hbar\, eB/mc = eV_y$. We multiply the above by e/h from both sides so that,

$$\frac{e^2 B}{2\pi mc} = \frac{e^2}{h} V_y \quad (2.5)$$

which describes the current in the x direction so that it is proved that, $\rho_{xy} = \frac{h}{e^2}$ which is the same as that required for the quantum Hall effect which uses only the Planck'sconstant and the charge of the electron. We take into account the gyromagnetic ratio so that (2.5) may be written as,

$$I_x = \frac{1}{2} g \frac{e^2 B}{2\pi mc} = \frac{1}{2} g \frac{e^2 V_y}{h}. \quad (2.6)$$

For $l = 0$, g=2,

$$I_x = \frac{e^2}{h} V_y \quad (2.7)$$

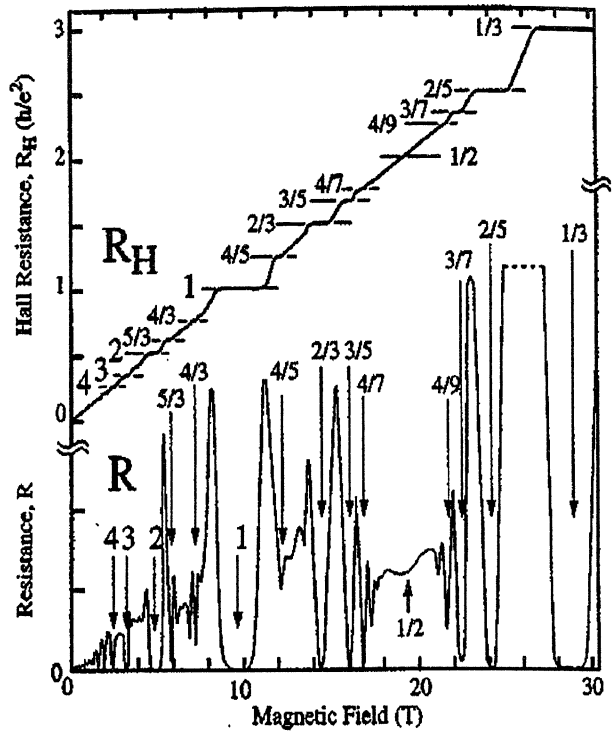

Fig. 2– The experimental fractional charges at which plateaus in the Hall resistivity occur. These fractions are the same as those calculated from the series given in Table. 1

which describes the quantized current correctly for ν = 1. From (2.6) $\nu = \frac{1}{2} g_\pm$ which gives the filling factor, one value for + sign and the other for – sign in (2.4). We have thus introduced three different g values, the usual g value as well as $g_\pm$. For l =0, we obtain $(1/2)g_+$=1 and $(1/2)g_-$=0, for l = 1, we get $(1/2)g_+$=2/3 and $(1/2)g_-$=1/3. These values of $\nu = (1/2)g_\pm$ are given in Table 1 from the theory of Shrivastava[5]. The predicted values are the same as those measured by Stormer as given in Fig. 2. The effective charge is determined by the modification of the cyclotron frequency, $\hbar\omega_c = g\mu_B B$ as,

$$e_{eff} = (1/2)g_\pm e = \nu e. \quad (2.8)$$

One of the series is $\nu_- = l/(2l+1)$ and the other is $\nu_+ = (l+1)/(2l+1)$. *These series exactly predict the series called Stormer-1.*

The formula can be rearranged as,

$$\frac{1}{2}g_{\pm} = \frac{l+\frac{1}{2}\pm s}{2l+1} \qquad (2.9)$$

For l=0, the above formula gives,

$$e^*/e = \frac{1}{2} \pm s \qquad (2.10)$$

Table 1– The calculated values of fractional charges for various values of l and s=±1/2.

S.No.	l	$(1/2)g_-=l/(2l+1)$	$(1/2)g_+=(l+1)/(2l+1)$
1	0	0	1
2	1	1/3	2/3
3	2	2/5	3/5
4	3	3/7	4/7
5	4	4/9	5/9
6	5	5/11	6/11
7	6	6/13	7/13
8	7	7/15	8/15
9	∞	1/2	1/2

so that for s=1/2, the effective charge becomes zero or one. Both these charges are important because they are both experimentally observed. We introduce the concept of negative spin to that the number of energy levels is not limited by $2S$+1. It does not matter much if this expression is relaxed. The number of levels will then be infinite and not $2S$+1. Upto 2x2 matrix representation for the spin, the commutators are the same for the negative spin as for the positive spin. In fact, the levels resemble the harmonic oscillator. In the case of oscillations in a many-body system, we can remove the divergence in the energy by fixing the number of atoms but in the case of spins, the equally spaced levels can be made to continue upto ∞.

First of all, we write the infinite set of energy levels as follows.

$$\frac{5}{2}g\mu_B H, \frac{3}{2}g\mu_B H, \frac{1}{2}g\mu_B H,$$

$$-\frac{1}{2}g\mu_B H, -\frac{3}{2}g\mu_B H, -\frac{5}{2}g\mu_B H, \ldots \qquad (2.11).$$

which are solutions of the Hamiltonian, $\mathcal{H}=g\mu_B H_z S_z$. Here g is the splitting factor, μ_B is the Bohr magneton, H_z the field along the z direction and S_z the z component of the spin. The above infinite series results for negative spin. For the positive spin, ½, the levels are at $+(1/2)g\mu_B H$ and $-(1/2)g\mu_B H$. Next, we use both the positive as well as the negative sign in the total angular momentum so that $j=l\pm$ s. In this case, we take the ratio,

$$g = \frac{2j+1}{2l+1}. \qquad (2.12)$$

Keeping both the signs in S, the above can be written as,

$$g = \frac{2(l\pm s)+1}{2l+1}. \qquad (2.13)$$

For positive sign,

$$\frac{1}{2}g_+ = \frac{l+\frac{1}{2}+s}{2l+1} \qquad (2.14)$$

and for negative sign,

$$\frac{1}{2}g_- = \frac{l+\frac{1}{2}-s}{2l+1} \qquad (2.15)$$

In the case of l =0, an interesting situation arises. The effective charge becomes related to spin,

$$e^*/e = \frac{1}{2}g_+ = \frac{1}{2}+s \text{ (Spin-charge locking)} \qquad (2.16)$$

and

$$e^*/e=\frac{1}{2}g_- =\frac{1}{2}-s \text{ (Spin-charge locking)} \quad (2.17)$$

Since, the Bohr magneton, $\mu_B=e\hbar$ /2mc multiplies the $g_\pm$ values, the effective charge of the electron can be written as $e^* = (1/2)g_\pm e$. For positive s=1/2, the equation (2.16) gives $g_+=2$ and for negative sign we get $g_-=0$. The $g_+=2$ is the usual expected value but we also find, $g_-=0$, which gives zero energy. We substitute $g=g_+=2$ (for s=1/2) in the expression (2.11) to obtain the following energies,

$$E=5\mu_BH,3\mu_BH,\mu_BH,-\mu_BH,-3\mu_BH,$$

$$-5\mu_BH \ , \ldots, \text{etc.} \quad (2.18)$$

and from eq.(2.17) we get, $g=g_-=0(s=1/2)$ which substituted in (2.11) gives,

$$E=0. \quad (2.19)$$

This zero is very important because it is associated with zero charge. We can eliminate μ_BH from the above by using,

$$g\frac{e\hbar}{2mc}H=\frac{g}{2}\hbar\omega_c=\hbar\omega_c \quad (2.20)$$

for $g=2$. For $s=1$, from (2.16) $g_+=3$, so we substitute this value in (2.11) to obtain,

$$\frac{15}{2}\mu_BH, \ \frac{9}{2}\mu_BH, \ \frac{3}{2}\mu_BH,$$

$$-\frac{3}{2}\mu_BH, -\frac{9}{2}\mu_BH, -\frac{15}{2}\mu_BH,\ldots,\text{etc.} \quad (2.21)$$

For $S=1$, with negative sign in front of spin, eq.(2.17) gives $g_-=-1$. We substitute $g=g_-=-1$ in (2.11) to obtain,

$$-\frac{5}{2}\mu_BH, -\frac{3}{2}\mu_BH, -\frac{1}{2}\mu_BH, +\frac{1}{2}\mu_BH,$$

$$+\frac{3}{2}\mu_BH, +\frac{5}{2}\mu_BH, \ldots, \text{etc.} \quad (2.22)$$

so that the positive and the negative energies get interchanged but there is no effect on the full energy level diagram. This is actually related to the invariance of the Hamiltonian with respect to the time reversal. For s=3/2, from eq.(2.16) $g_+= 4$. We substitute $g = g_+= 4$ in eq.(2.11) to obtain,

$$10\mu_BH, 6\mu_BH,2\mu_BH, -2\mu_BH,$$

$$-6\mu_BH, -10\mu_BH, \ldots, \text{etc.} \quad (2.23)$$

so that we obtain the series,±2, ±6, ±10, ±14, ±18, etc. This series has the interval 4. Not only the interval but the full series is the same as in the experimental data[16] at 9 Tesla as shown in Fig. 3. We have obtained the exact series without any approximation and all numbers are individually exact. Hence, the many-body corrections are not the cause of the series. For $s=3/2$, using the negative sign expression (2.17) we obtain, $(1/2)g_-=-1$. In this case, the energy levels of $g=-2$ are the same as that of $g=2$ due to time reversal invariance. If there are N atoms of spin each equal to ½, the total spin of N particles in $N/2$. The energy levels of the type (2.11) will be found. As the magnetic field is varied, different energy levels cross the Fermi level so that there are oscillations but that will not form plateaus. The plateaus are formed due to the flux quantization,

$$\pi l_o^2 B=n\,\phi_o \quad (2.24)$$

where $\phi_o=hc/e$. We substitute the flux quantization condition in eq.(2.11) so that the magnetic field completely disappears,

$$\frac{5}{2}g\mu_B(\frac{n\phi_o}{\pi l_o^2}), \ \frac{3}{2}\ g\mu_B(\frac{n\phi_o}{\pi l_o^2}), \ \frac{1}{2}\ g\mu_B(\frac{n\phi_o}{\pi l_o^2}),$$

$$-\frac{1}{2}g\mu_B(\frac{n\phi_o}{\pi l_o^2}),-\frac{3}{2}g\mu_B(\frac{n\phi_o}{\pi l_o^2}),\ldots, \quad (2.25)$$

Not only that there are factors like, 5/2, 3/2, 1/2, -1/2, -3/2, -5/2, ..., ∞, there is a factor of n also which has come from the flux quantization. We

substitute the flux quantization in the series (2.23) so that we find,

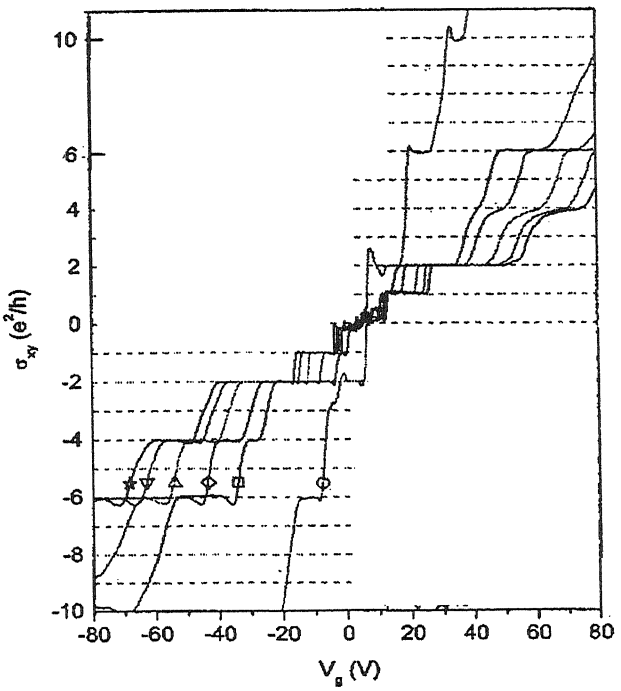

Fig. 3– The Hall conductivity of a graphene sample at a magnetic field of 9Tesla(circle), 25T(square), 30T(diamond), 37T(up triangle), 42T(down triangle) and 45T(star).

$$10\ \mu_B(\frac{n\phi_o}{\pi l_o^2}),\ 6\ \mu_B(\frac{n\phi_o}{\pi l_o^2}),\ 2\ \mu_B(\frac{n\phi_o}{\pi l_o^2}),$$

$$-2\ \mu_B(\frac{n\phi_o}{\pi l_o^2}),-6\ \mu_B(\frac{n\phi_o}{\pi l_o^2}),$$

$$-10\ \mu_B(\frac{n\phi_o}{\pi l_o^2}),\ldots, \quad (2.26)$$

The series for n=1 is ..., 10, 6, 2, -2, -6, -10, ... but for n=2 we have ..., 20, 12, 4, -4, -12, -20, ..., ∞. Now the interval is not 4 but 8. The plateaus are now predicted at ±4 which was not a part of the first series. When the field is increased from 9 Tesla to 25 Tesla the plateaus at ±4 are indeed shown in Fig. 3. The remaining values are too weak. In this way we are able to predict the plateaus at ±4 in addition to those given by the series 2, 6, 10, ..., etc. This means that our predicted values are in full agreement with the data. The values of 0 and 1 were already present in earlier work[5] as seen in Table 1. Away from the plateau the field dependence is of the form of, $10\ \mu_B(\frac{n\phi_o}{\pi l_o^2}) + 10\mu_B H$ as an example, which is $a_1+a_2H=a_3H^{1/2}$. Now the Hamiltonian will be of the form, $g_+\mu_B H + g\text{-}\mu_B H$ so that there are two terms instead of the usual one. The 4x4 Dirac matrices are obtained for only one Zeeman Hamiltonian. Then using two types g values will result into 8x8 Dirac matrices. The symmetry of the series (2.14) and (2.15), also given in Table 1 is the same as in the experimental data[3,17] as shown in Fig. 4. The symmetry properties are also given in ref. 10.

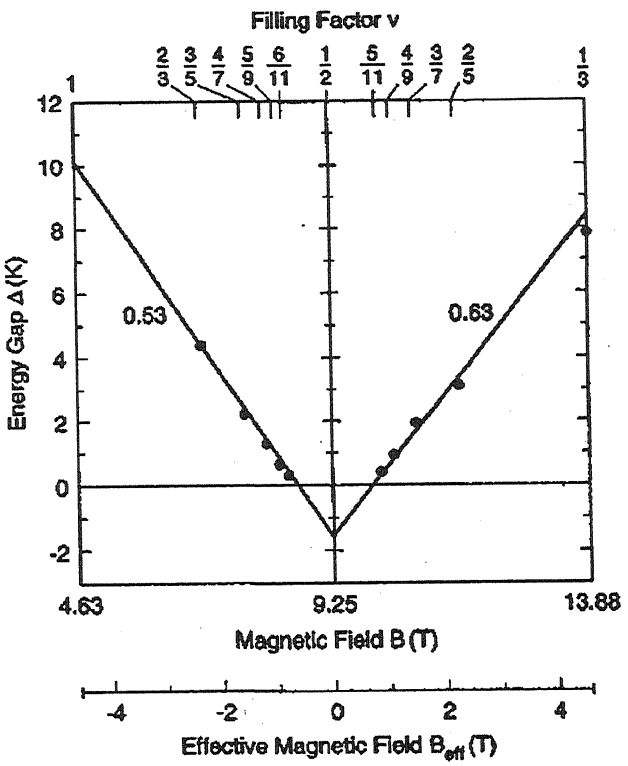

Fig. 4– The experimental data showing the same symmetry as predicted.

Spin-Charge Locking

In Laughlin's theory, the fractional charge is generated due to flux quantization and incompressibility due to correlations amongst electrons but without the use of spin. In the present theory, the spin of the electron occurs in the g value

via the ratio of the total angular momentum to the orbital angular momentum. Later on, the cyclotron frequency is modified to $(g/2)\hbar\omega$ which is equivalent to multiplying the Bohr magneton as $(g/2)e\hbar/mc$. Hence, it is possible to define the charge of the electron as $e^*=(g/2)e$. If the charge is treated as a vector, then it is possible to align a component of the charge with a component of the spin so that spin and charge get locked. When spin changes, there is a change in the effective value of the charge which can be determined from the resistivity,

$$\rho = \frac{h}{ie^2} \qquad (2.27)$$

Now, i is equal to ±2, ±6, ±10, ±14, ..., etc. We say that the effective charge of the electron becomes, $e^*= \pm 2e, \pm 6e, \pm 10e, \pm 14e,...$, etc. The charge is clearly determined by spin as in (2.16) and (2.17). Therefore, there is a spin-charge locking. The Chern numbers are the random numbers which depend on the topology of the sample. In the case of the present series, not only that there is an interval of 4 but also the values are definite and exact. Hence they are unlikely to be "Chern " type.

Conclusions

The predicted series of fractions of the electron charge such as 1/3, 2/3, ..., given by Table 1, is correct and agrees with that experimentally measured by Stormer. Similarly large charges such as 2, 6, 10, ..., predicted by our theory are in agreement with those measured in graphene. We learn that there is strong spin-charge effect. In the quark model, the hypercharge, Y, is given by baryon number, B and strangeness, S as $Y=S+B=2(Q-1)$ where Q is the charge, which does not use spin and is applicable to the hadrons only. In our case, the electron charge splits into many quasiparticles such that the charge depends on spin. Large charges are also found. In superconductivity[19] it is sufficient to make Cooper pairs. However, in the present case there are both spin up and down states with much more involvement of spin than in the superconductors. The present theory has serious effect on the charge and the mass of the electron[20-24].

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Effect of ammonia inhalation on serum protein of Albino rat

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Abstract

Ammonia is a strongly alkaline chemical which is widely used in industry as a feed stock for nitrogen based chemicals such as fertilizers, explosives and plastics. In the present study albino rats were exposed to 50 and 150 ppm of ammonia gas to study the toxic effect of ammonia on serum protein level of exposed rats.

Keywords : ammonia, protein, Albino rats.

Introduction

The gaseous pollutants adversely affects the living species since they are capable of penetrating the defence mechanism of the nose and upper air passage through nasopharyngeal, tracheobranchial and pulmonary compartment and can reach the smaller passages affecting respiratory system and blood[1]. Ammonia and various nitrogen oxides (NO_x) are two nitrogen compounds of particular environmental concern. Ammonia is a part of nitrogen cycle and is present in the environment as a result of natural processes and industrial activity, including certain types of intensive farming, mostly come from agricultural activities. About 260,000 tones of nitrogen are emitted each year as ammonia[2]. Ammonia gas releases heat as it dissolves in water and can cause thermal injury which can cause changes in serum activity of some enzymes. This paper shows that ammonia inhalation by rats results in the decrease of serum protein in them.

Material and Methods

The serum total protein was determined by Biuret method.

Procedure

Three test tubes marked as Test 'T', Standard 'S' and Blank 'B' were taken.

1.0ml of Biuret reagent was added in each test tube, 0.02 ml of serum sample was added to test tube marked T and S while in B distilled water was added instead of serum. Mixed well and incubated at 37°C for 5-10 minutes.

The optical density of test and standard was measured against blank at 546 nm using visual spectrophotometer (Spectronic). The Protein content can be calculated by using the following formula.

Serum total protein (gm / dl) =

$$\frac{O.D\ of\ Test}{O.D\ of\ \text{Standard}} \times 8$$

Results and Discussion

The total serum protein of control rats after 15 days ranges from 6.00 to 7.87 gm/dl with an average of 6.64 gm/dl whereas the total serum protein after 15 days exposure to 50 ppm ammonia ranges from 5.21 to 6.92 gm/dl with an average of 5.8 gm/dl and after 15 days exposure to 150 ppm ammonia ranges from 5.01 to 6.32 gm/dl with an average of 5.56 gm/dl. The decrease in the mean value of total serum protein is very highly significant after 15 days exposure after 15 days exposure to 150 ppm NH_3 gas (Table 1 and Fig 1).

The total serum protein of control rats after 30 days ranges from 6.17 to 7.65 gm/dl with an average of 6.69 gm/dl whereas the total serum protein after 30 days exposure to 50 ppm ammonia ranges from 5.15 to 5.86 gm/dl with an average of 5.46 gm/dl) and after 30 days exposure to 150 ppm ammonia ranges from 4.89 to 5.95 gm/dl with an average of 5.20 gm/dl.

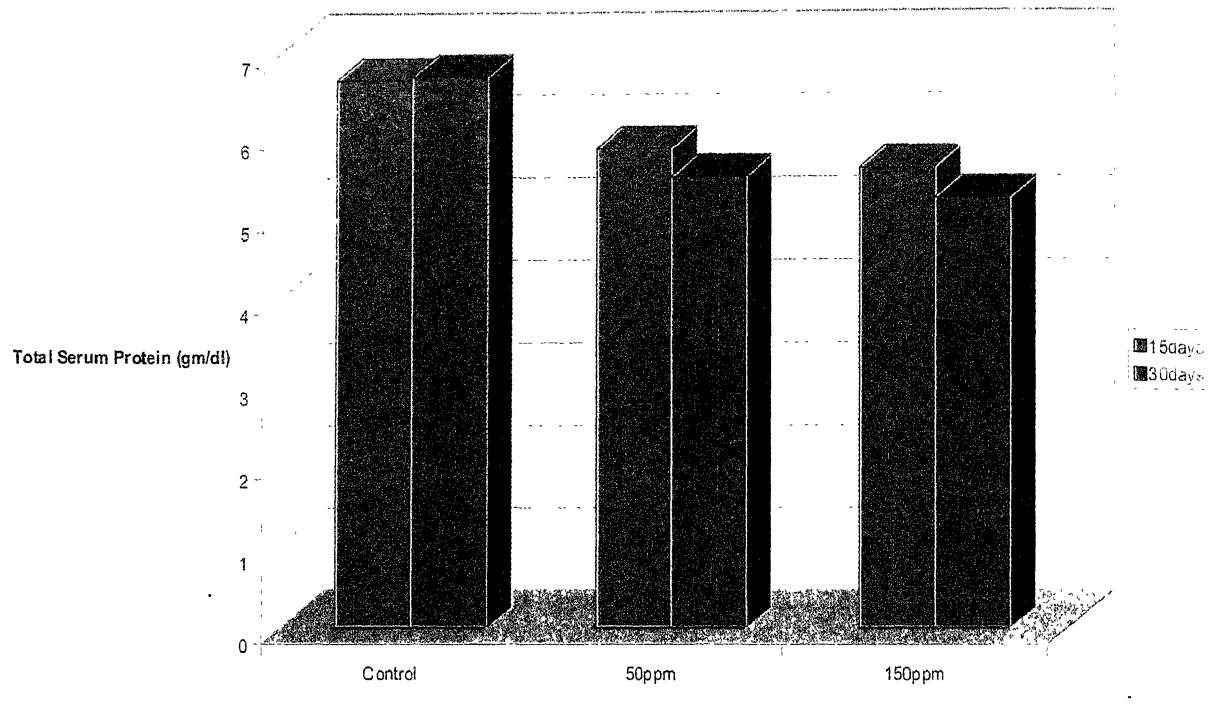

Fig. 1- Total Serum Protein (gm/dl) in albino rats after exposure to 50 and 150 ppm

Table 1- Total serum protein (gm /dl) in albino rats after exposure to NH_3

No. of Rats	Exposure Day	Control Set	Experimental Set	
			NH_3 Concentration	
			50 ppm	150 ppm
		Range Mean ± S.Em	Range Mean ± S.Em	Range Mean ± S.Em
5	15	6.00 - 7.87 (6.64 ± 0.354)	5.21 - 6.92**** (5. 8 ± 0.312) ↓	5.01 - 6.32**** (5.56 ± 0.328) ↓
5	30	6.17 - 7.65 (6.69 ± 0.321)	5.15 - 5.86*** (5.46 ± 0.136) ↓	4.89 - 5.95**** (5.20 ± 0.196) ↓

Decrease
ppm- parts per million
SEm standard error of mean

*** Highly Significant (P <0.01)
**** = Very Highly Significant (P <0.001)

The decrease in the mean value of total serum protein is highly significant after 30 days exposure to 50 ppm whereas after 30 days exposure to 150ppm NH_3 gas the decrease is very highly significant as given in Table 1 and Fig 1.

Enzymes are proteins which are present as globulin portion of serum. They are not confined solely to the serum but are present in various portions of cells, though the amount in different organs can vary widely. When an organ is damaged or cell gets injured, a greater amount of enzyme leaks out into plasma. The extent of the rise or fall in serum activity of these enzymes depends on the concentration of enzymes in the tissue and on the severity of damage. The rate at which an enzyme leaks from damaged tissue is affected by location of the enzyme in the cells and by change in the permeability of the cell membrane.

The decrease in total protein level is due to respiratory inflammation in albino rats which accompanies epithelial cell injury due to inhalation of ammonia and results in increase of epithelial and capillary membrane permeability which causes leakage of proteins from serum to site of pulmonary injury which leads to the decrease in total protein level value in albino rats. Some workers have also reported decrease in protein due to exposure of oxides of nitrogen[3,4,5,6,7]. NH_3 form ammonium ions which enter the liver via portal vein and are converted to urea and higher ammonia concentration can results into protein deamination[8,9,10].

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A brief report on the Symposium on "Novel approaches for food and nutritional security" and 77th Annual Session of the Academy, held at CFTRI, Mysore from December 6 – 8, 2007

Symposium : After independence efforts have been made to strengthen and sustain the food and nutrition security. However, there remains a lot to be done to enhance the scientific and technical capabilities, sharpen the knowledge base and expertise towards meeting the challenges of food and nutritional security. The success towards meeting these challenges is dependent more on coordination and harmonization of the scientific and technological leads achieved towards feasible implementation approaches. There is a growing recognition among institutions engaged in these efforts to bring in high scientific caliber and technical skills through a holistic approach. The final goal should be to benefit the society and improve the quality of life. Realizing this, The National Academy of Sciences, India (NASI), Allahabad, in association with Central Food Technological Research Institute (CFTRI), Mysore, organized a symposium on "Novel approaches for food and nutritional security" during its 77th Annual Session held at CFTRI, Mysore from December 6 – 8, 2007. Dr. (Mrs.) Manju Sharma, the Past President of the Academy, took the lead (as Convener) in organizing this Symposium.

The Symposium and 77th Annual Session were inaugurated by Hon'ble Prof. M.G. K. Menon, FRS, Advisor, Indian Space Research Organization, Govt. of India and Past President of the Academy on December 6, 2007. Prof. Menon while emphasizing that food and nutritional security depends on multiple variables, stressed on proper utilization of advancement in biotechnology, agricultural science and food research to combat malnutrition and related diseases/ disorders in large Indian population. His inspiring speech was in consonance with the ideas of Her Excellency Mrs. Pratibha D. Patil, President, Republic of India, who expressed in her message- "As an Academy concerned with how science can benefit society, this year's annual session and symposium will, I am confident, deliberate on societal awareness and novel scientific approaches towards combating malnutrition particularly amongst women and children". Prof. Menon further said that academies and scientists should not only think of themselves and science, but also reach out to all so that even the disadvantaged section would benefit from science and technology. Pointing out that a large number of people still lived below the poverty line and did not get food and nutrition or education, Prof. Menon said that it was the duty of scientists to reach out to such people. "I am not bothered about reservation in IITs and IIMs as it is trivial and not the solution," he said and called for a fundamental change in attitude to conceive an all inclusive policy to bring the poor and the disadvantaged section into the domain of science that was not superficial but was characterized by depth.

Refuting the notion that science was something pure only in pursuit of truth and was not relevant to daily life, Prof. Menon gave examples of the relevance of science in day to day applications and pointed out that the recent Nobel Prize for Physics was awarded for breakthrough in nano-technology which had application in computer hard discs. Recalling the days when a large congregation of people would gather to listen to scientists, Prof. Menon noted that in contrast the venue of the programme in Mysore was not full and this was reflective of the state of science in general and exhorted scientists and teachers to make the subject exciting.

The technical session began with the address of Dr. V. Prakash, Director, CFTRI, Mysore on " A vibrant Indian economy through a vibrant nutritional security – A long term investment for reaching our science to society". Dr. Prakash dealt in detail about nutritional security in our nation with the back drop of food science, food technology and agriculture; and quoted " What is needed is healthy long life and certainly not just long life alone", with a view to attain adequate nutrition to all by 2012, an optimistic target for India. Dr. B. Sesikeran, Director, National Institute of Nutrition, Hyderabad delivering an informative address on " The current nutritional situation in India" further analyzed and strengthened the views of Dr. Prakash. The Chairpersons of this opening technical session were Prof. Ashok Misra, President, NASI & Director Indian Institute of Technology Bombay, and Prof. B.N. Dhawan, Former Director, Central Drug Research Institute, Lucknow.

The technical session-II was focused on "Enhancing food and nutritional security – Research and development". Dr. Kamla Krishnaswami, Former Director, National Institute of Nutrition, Hyderabad initiated the discussion on this theme in her address on " Vegetables and fruits as modern miracles for malnutrition – Diet related chronic disorders". She expressed that the scientific basis supporting the extraordinary health benefits obtained by eating vegetables and fruits is growing rapidly. Vegetables and fruits appear to be playing a prominent role in prevention of several diet related chronic diseases. The earlier research studies show that these dietary items contain a variety of nutrients, phytochemicals and fiber which impact micronutrient deficiencies, the aetiopathogenisis of cardiovascular diseases, cancer, cataract, osteoporosis, diabetes, chronic obstructive pulmonary diseases and diverticulosis. Their consumption will significantly affect the life span and quality of life. Promoting healthy dietary patterns and eating habits are challenging tasks to improve nutritional status with respect to both micro and phyto nutrients. Prof. Akhilesh K. Tyagi, Director, Inter- disciplinary Center for Plant Genomics, University of Delhi, South Campus, New Delhi in his address on "Promise of rice biotechnology in addressing food and nutritional security" suggested that remedy to food shortage could be a better management of seed/ hybrid seed supply, sensitization of farmers to new farm technology and cooperative/ corporate farming. Prof. Tyagi further analyzed the role of rice biotechnology in meeting the increasing demand of food due to population boom. The last address of this session was delivered by Ms. Ana Abraham Sinha, Head,

Corporate Wellness and Consumer Service, Nestle India Ltd., Gurgaon on "Nutrition, health and wellness – A way forward". While detailing the involvement of Nestle in providing consumers the relevant information on improving the quality of diet and thereby life, she gave a comparative account of the emergence of life style related diseases and a growing awareness towards nutrition, health and wellness. The session was chaired by Dr. Vijayalakshmi Ravindranath, Director, National Brain Research Center, Manesar, Haryana and Dr. P.V. Salimath, Head, Department of Biochemistry and Nutrition, CFTRI, Mysore.

After an overview of the research and development, the third session discussed the technical developments and their adaptation/ transfer, coordinated by two well known experts Dr. V.P. Kamboj, Former Director, Central Drug Research Institute, Lucknow and Dr. Rakesh Tuli, Director, National Botanical Research Institute, Lucknow as the Chairpersons. Dr. G.S. Kalloo, Vice Chancellor, Jawaharlal Nehru Krishi Vishwavidyalaya, Jabalpur, emphasized on "Biotechnology and productivity enhancement: Current status and future strategy". He was of the opinion that research on genomics and proteomics could bring out a major change in solving many biological problems. Dr. A.S. Bawa, Director, Defence Food Research Laboratory (DFRL), Mysore gave an overview on " Food Technologies Development at DFRL". One of the most important technologies developed by different food research laboratories is ready–to-eat products stabilized by antimycotic agents, retort pouch processing technology, minimal processing of vegetables, intermediate and hurdle technology fruits, self heating systems for compo pack rations, shelf stable tender coconut water, etc. Dr. Ajay Parida, Programme Director, M.S. Swaminathan Research Foundation, Chennai gave a detailed account of "Biotechnology option for enhancing food and nutrition security"; especially the role of M.S.S.R.F. in contributing the know-how for the enrichment of genetic diversity of crops and others plants.

The technical session-IV started on 7th December on a theme- " Nutritional Outreach". Dr. Mahtab S. Bamji, Dangoria Charitable Trust, illustrated several methods for " Nutritionally and environmentally promotive farming", with the active involvement of women as the major stake holders. Dr. Jamuna Prakash, Department of Food Sciences & Nutrition, University of Mysore spoke on " Addressing food and nutrition security in urbanization India-Issues, concerns and interventional approaches", stressing upon pragmatic steps including policy reform, collective social action and change in supply system as remedial measures for the nutritional insecurity among the urban poor. The session was Chaired by Prof. J.P. Mittal, Former Director, Chemistry & Isotopes Groups, Bhabha Atomic Research Center, Mumbai and Prof. G. Saraswathi, Department of Food Sciences & Nutrition, University of Mysore.

Finally, the session on food safety and security was addressed by Dr. Akshay K. Pradhan, Tata Energy Research Institute, New Delhi and Dr. Anupa Siddhu, Director, Lady Irwin College, New Delhi on

"Breeding of mustard for better nutrition" and "Awareness: Technologies", respectively. The session was Chaired by Dr. S.P.S. Khanuja, Director, Central Institute of Medicinal and Aromatic Plants, Lucknow and Dr. A.G. Appu Rao, Head, Protein Chemistry & Technology, CFTRI, Mysore.

The concluding session was Chaired by Dr. (Mrs.) Manju Sharma. The other distinguished scientists present were Dr. Kamala Krishnaswami, Dr. Mahtab S. Bamji, Dr. V.P. Sharma, Dr. Rakesh Tuli, Dr. Akhilesh K. Tyagi and Dr. P.K. Seth, who participated actively in the discussion.

Dr. (Mrs.) Manju Sharma gratefully acknowledged the contributions of all the eminent speakers and thanked them for their valuable suggestions. She was deeply concerned about the poor nutritional status of a large number of Indian populations, especially the women. Dr. (Mrs.) Sharma expressed that from the deliberations of the symposium it could be inferred that problem lies at different levels, but solutions are also in sight.

After a thorough discussion the following recommendations were made –

1. There is a need to enhance the crop yield to ensure adequate supply of essential food grains, vegetables and fruits. Indian Council of Agricultural Research, Horticulture Department, Department of Biotechnology and other such agencies should take responsibilities to invent methods for enhanced production and proper implementation of lab to land programs. There is a need to integrate biotechnological tools with molecular breeding and selection of germplasm for enhancing the quality of food items.
2. Mass awareness campaign, rather a movement, need to be launched to spread the know-how of cutting-edge technology for sustainable agricultural production and quality improvement in food and food products.
3. Further, science of nutrition should reach society to ensure not only the appropriate consumption of energy rich food but also protective food, as well. For this awakening and educational programs at community level be strengthened with special emphasis on the role of women for effective nutritional intervention.
4. The higher incidence of low birth weight babies and micronutrient deficiency diseases be checked by adequate supply of nutrient rich food and food products as well as educating communities at different levels, especially the women.
5. Mid day meal to be provided in all the schools on low cost (as Rs. 10/child/day) with the intention of its nutritious values. Home gardens and school gardens be setup to ensure regular intake of fresh vegetables for the children.
6. NGOs and local bodies (like Panchayat, Seva Samiti etc.) be actively involved to create nutritional awareness and train the common mass at grass- root level.

7. Food adulteration must be checked by strict supervision involving governmental and non-governmental agencies and mass awareness against the menace of adulterated food.

8. As an outreach, nutritional education be integrated as curriculum in the graduation and post graduation science courses, especially in agricultural and medical sciences.

9. Further, to strengthen the knowledge base and transfer technologies a chain of trained professionals be created at different steps in the society to develop a network with the help of ICAR, Academies, Universities and other bodies.

10. A large-scale publicity of the network and nutrition enrichment programs be ensured with the help of Doordarshan, ISRO transponders and other channels supported by industries and governmental agencies.

11. The National Academy of Sciences, India, Allahabad may prepare a policy to combat malnutrition, especially among the women and children in India, so that socio- economic sustainable development be ensured

12. And lastly, health and nutrition should be the yardstick of national development.

It was unanimously resolved that the above recommendations would be communicated to the Government for inclusion/ implementation during the 11^{th} plan to change the scenario drastically for a healthy nation.

Seventy-seventh Annual Session: The Seventy-seventh Annual Session of the Academy included Addresses of the President, Sectional Presidents, Invited Speakers and presentation of research papers by the scientists including Fellows and Members of the Academy on December 6-8, 2007. Several prestigious awards were also given during the Session.

Prof. Ashok Misra, President of the Academy while delivering the Presidential Address on 'Developments in Polymers' in the Inaugural Session, remarked that polymerization is the biggest contribution of chemistry in the 20^{th} century. He enumerated the increasing usage of plastic in various fields. He stated that the use of plastic in agriculture and food industry has fuelled growth; and that plastic is replacing load bearing components in several engineering applications as well. He explained that plastic is being used predominantly in automotive industry, railways, aerospace industry, medical applications, health care, structures, marine applications, sports industry, transportation, textile, electronics, packaging and bone replacement too.

Prof. Misra also expressed his happiness on the level rise in the stature of NASI during the last few years; but simultaneously stressed on not to forget the objective i.e. "to impart cultural improvement by contribution to human knowledge".

The Presidential Addresses in the Sections of Physical and Biological Sciences were delivered by Prof. S.C. Dutta Roy, Emeritus Professor, Department of Electrical Engineering, Indian Institute of

Technology, Delhi and Prof. Pramod Tandon, Vice Chancellor, North Eastern Hill University, Shillong, respectively, on December 7, 2007. Prof. S.C. Dutta Roy while delivering his address on "In praise of teaching and research as a career option" emphasized the role of teaching and research in shaping the society; as well as highlighted the ethical values and honour associated with the profession. Prof. Tandon through his illustrative address focused on conservation of plant diversity with the help of molecular tools and biotechnological advancements to make it rich and prosperous. Both the addresses were inspiring for the young researchers giving a boost to their zeal for opting science as a career in future.

In both Sections (physical and biological) invited talks were also delivered. The invited speakers in Physical Sciences Section were Prof. B.K. Roy, Indian Statistical Institute, Kolkata, Dr. G.V.M. Sharma, Indian Institute of Chemical Technology, Hyderabad and Prof. B.L. Deekshatulu, University of Hyderabad, Hyderabad; and in Biological Sciences Section invited speakers were Prof. T. Mohapatra, Indian Agricultural Research Institute, New Delhi, Prof. P.N. Rangarajan, Indian Institute of Sciences, Bangalore, Dr. Soniya Nityanand, S.G.P.G.I, Lucknow and Prof. N.K. Chrungoo, North Eastern Hill University, Shillong.

300 research papers were presented in both the Sections. NASI- Swarna Jayanti Puraskars for best research paper presentations were given to Dr. Ms.V. Radhika Devi, School of Physics, University of Hyderabad, Hydrabad and Dr.Ms. Shreemoyee Bordoloi, Department of Chemistry, Dibrugarh University, Dibrugarh in the section of Physical Sciences; and to Dr.Ms.Shritapa Datta, Biochemistry Adaptation Laboratory, Department of Zoology, North Eastern Hill University, Shillong and Dr.Ms.Nimisha Kankan, Department of Zoology, Deendayal Upadhyay Govt. Degree College, Saidabad, Allahabad in the section of Biological Sciences.

Earlier, NASI– Reliance Industries Platinum Jubilee Awards (2007) for application oriented innovations covering both Physical and Biological Sciences were given to Prof. Bimal Kumar Roy, Dean of Studies, Indian Statistical Institute, Kolkata and Dr. G.V. Madhava Sharma, Scientist- F (Deputy Director), Discovery Laboratory, Indian Institute of Chemical Technology, Hyderabad (in Physical Sciences) and Dr. P.N. Rangrajan, Associate Professor, Department of Biochemistry, Indian Institute of Science, Bangalore and Dr. Tirlochan Mohapatra, Principal Scientist, NRC on Plant Biotechnology, Indian Agricultural Research Institute, New Delhi (in Biological Sciences).

During the Fellows' Meeting, held on December 7,2007 besides other deliberations, those Fellows who had not yet signed the Fellow's register, put in their signatures. In the Annual General Body Meeting, also held on December 7, 2007,besides other deliberations the name of NASI- Senior Scientist Platinum Jubilee Fellows (2007) viz. Prof. Suresh Chandra, Physics Department, Banaras Hindu University, Varanasi, Prof. J.S. Singh, Botany Department, Banaras Hindu University, Varanasi and Prof. S.P. Vij, Botany Department, Panjab University, Chandigarh were also announced.

The Valedictory Function was presided over by Prof. Ashok Misra, President of the Academy. The Sectional Presidents and General Secretaries were also present on the occasion. Prof. Ashok Misra, President of the Academy presented NASI- Young Scientist Platinum Jubilee Awards (2007) in the filed of Biological, Physical and Chemical Sciences to Dr. Rajeev K. Varshney, Senior Scientist, Applied Genomics Laboratory, International Crops Research Institute for the Semi- Arid Tropics, Patancheru, Hyderabad, Dr. Gopaljee Jha, Centre for Cellular and Molecular Biology, Hyderabad, Dr. Lily Goswami, JRF, Bose Institute, Kolkata, Dr. Mohan Mondal, Scientist, National Research Centre on Mithun, Nagaland, Dr. Deepak Narhari Modi, Sr. Research Officer, National Institute for Research in Reproductive Health, Mumbai, Dr. Subhabrata Chakrabarti, Staff Scientist, L.V. Prasad Eye Institute, Hyderabad, Dr. Suman Chakraborty, Assistant Professor, Department of Mechanical Engineering, Indian Institute of Technology, Kharagpur, Dr. Deepankar Choudhary, Assistant Professor, Department of Civil Engineering, Indian Institute of Technology Bombay, Mumbai, Dr. Manoj Kumbhakar, Scientific Officer D, Radiation and Photochemistry Division, Chemistry Group, Bhaba Atomic Research Centre, Mumbai and Dr. (Ms.) Suman Lata Jain, Research Associate, Process Engineering Applied Chemistry and Biotechnology Division, Indian Institute of Petroleum, Dehradun.

Prof. P.K. Seth, General Secretary (HQ), expressed vote- of- thanks at the end of the Session.

A Report on Science Communication Programme:-

I. Science Extension Lectures: The following lectures were held:-

Outstation

A. The speakers Dr. Pankaj Srivastava (on Cyber Crime), Dr. Md. Masood (on Food Adulteration), Dr. Deepak Chandra (on Biotechnology) and Dr. Kamlesh Pandey (on artificial diamonds) delivered lectures at BRP Inter College, T.D. Inter College and Janak Kumari Bal Shiksha Vidyalaya, at Jaunpur on January 24 & 25, 2008.

B. Dr. Rajeev Srivastava delivered lecture on computer education at St. Andrew's Inter College, Gorakhpur on January 25, 2008.

In Allahabad

Date	Venue	Topic	Speaker
1. Feb. 9, 2008	NASI	Prof. U.S. Srivastava Memorial Award Lecture on 'Educational Philosophy of Biology'	Prof. K. Muralidhar
2. Feb. 11, 2008	NASI	Biodiversity Climate Change	Prof. J.S.Singh Prof. S.L. Srivastava
3. Feb. 17, 2008	Botany Dept., A.U.	Chronological details of Research in Photosynthesis.	Prof. Govindjee
4. Feb. 19, 2008	NASI	Photosynthesis in our life	Prof. Govindjee

II. Winter School

The National Academy of Sciences, India organized a Winter School for the first time under its Science Communication Programme from February 15 to 17, 2008 at Biotech Park, Lucknow. The DNA Club Members also participated in this school. 82 teachers and 220 students of +2 level from 30 Districts of U.P., M.P. and Bihar participated in the school. The inaugural function was held on 15th February, 2008 in Biotech Park, Lucknow with Hon'ble Sri Abdul Mannan, Minister, Science and Technology, Govt. of Uttar Pradesh as the Chief Guest. Prof. P.K. Seth, General Secretary, NASI & CEO, Biotech Park welcomed Hon'ble Sri Mannan, other distinguished guests, teachers and student-participants. On this occasion, Dr. Rakeh Tuli, Director, NBRI and CDRI, Lucknow gave a brief account of the depleting natural resources and Prof. C.B.L. Srivastava emphasized the importance of natural resources and its conservation. Sri Ram Kumar, Principal Secretary, Science & Technology Department, Govt. of U.P., presided over the function. Dr. Niraj Kumar proposed the vote-of-thanks.

After the inaugural function all the participants visited the Botanical Garden of National Botanical Research Institute observing the diversity of plant species. From there, they went to the Central Institute of Medicinal and Aromatic Plants (CIMAP) to learn the techniques of conservation of plant resources and interact with the eminent scientists and researchers. In the evening, lectures were delivered by Prof. S.P.S. Khanuja, Director, CIMAP and Prof. Veena Tandon, North-Eastern Hill University, Shillong in the auditorium of CIMAP. Prof. Khanuja emphasized the chronological advancements in genetics and molecular biology, while Prof. Veena Tandon dealt in detail about the types of worms and their association with the animals and human being. In the evening of 15th a nice cultural evening was enjoyed; the programme was organized by the artists of Regional Office, *Geet Evam Natya Prasar,* Lucknow, Ministry of Information & Broadcasting, Govt. of India. at Biotech Park, Lucknow.

On February 16, 2008 the participants went to Biotech Park to visit Bioinformatics lab, Tissue Culture lab and DNA Bank. After the lunch they visited Regional Science Center, where they performed few experiments. In the evening of 16th Prof. R.S. Nadgauda, Indian Institute of Advanced Research, Ghandhinagar and Prof. Paramjeet Khurana, Delhi University delivered illustrative lectures on Bamboo and Biotechnology, respectively.

On February 17, 2008 arrangement for the site seeing (*Imambara and Bhulbhullaya*) was made and after that, the participants visited Birbal Sahni Institute of Palaeobotany to learn many things about palaeontology and its methods. The afternoon of this day was exciting for the students as they saw several endangered species in the zoo.

Certificates of participantion were given to all the teachers and students who attended the school.

III. National Science Week & National Science Day- 2008

The National Academy of Sciences, India organized the Science Week from February 20 to 27, 2008 on the eve of National Science Day (February 28, 2008). Earlier, the Academy also successfully organized Allahabad District Level Talent Search Examinations (Physical Sciences, Biological Sciences and Chemical Sciences). From this year, a National Scientific Writing Contest for the undergraduate students was organized, in which 72 students participated, from all across the country. Science Quiz, Debate, Oration, Exhibition, Creative Writing and Painting Contests and also two Workshops, one on Physics and the other on Health, were organized during the Science Week. Local Level Science Communication Activities were held from 20th February to 23rd February, 2008; and National & State Level Science Communication Activities started from 24th February 2008, inaugutated by Dr. Anil Kumar, NCL, Pune who delivered N.R. Dhar Memorial Lecture on "Physical Forces Responsible for Realizing Reactions of Organic and Biological Relevance". Different activities were held from 24-27 February, 2008 and the Science Day was celebrated on 28th February, 2008 **In total about 10,000 students participated in District and State Level Contests.**

The Gold-Medals were won by the following:-

1. Prof. H.C.Khare Memorial Gold Medal -2008 – Ms. Mehwish Khan, Buland Sahar
2. Prof. U.S. Srivastava Memorial Gold Medal -2008 –Mr. Ankur Gupta, Etawah
3. Prof. Krishnaji Memorial Gold Medal -2008 –Ms. Bharti Kumari, Agra

The winners of National Scientific Writing Contest are:-

1. Mr.Tirtha Banerjee, Jadhavpur University, Kolkata
2. Ms.Gitali Sharma, ITS Paramedical Sciences, Muradnagar, Ghaziabad
3. Ms.Jyoti Jain, Govt. Medical College, Aurangabad

Dr. Upkar Dutt Sharma, Meerut was awarded The National Academy of Sciences, India- Science Teacher Award for the year 2008.

Topics for the Debate, Oration and Scientific Writing Contests were:-

Debate: "Is Nuclear Deal with USA in our interest?"
Oration: "Pollution risks to health".
Scientific Writing Contests: "Climate Change".

Prof. Abhai Mansingh, formerly Director of South Campus, Delhi University was the Chief Guest on the Science Day. Prof. Mansingh delivered a talk on 'Science and Technology- Future Job Scenario'. He was of this opinion that the nation to compete with the developed countries, science and technology should be developed such that basic science is not ignored. Earlier Prof. S.L. Srivastava gave a brief account of all the activities organized during the year 2007-08 with the help of more than 50 resource persons. Prizes & Certificates were also given to the winners by Prof. Mansingh. Prof. U.C. Srivastava proposed the vote-of-thanks.

All these activities were organized under overall supervision of Prof. S.L. Srivastava, Coordinator, Science Communication Programme of the Academy. The scientists who conducted the various activities were:- Prof. Govindjee (U.S.A.), Prof. C.B.L. Srivastava, Dr. Abhai Mansingh, Prof. J.S. Singh, Prof. H.C. Verma, Dr. Anil Kumar, Prof. P.K. Seth, Prof. S.P.S. Khanuja, Prof. Veena Tandon, Prof. Paramjeet Khurana, Prof. R.S.Nadgauda, Dr. Rakesh Tuli, Prof. M.M. Laloraya, Prof. G.K. Srivastava, Prof. C.K. Dwivedi, Prof. U.C. Srivastava, Prof. Pratima Gaur, Prof. Anita Gopesh, Prof. Krishna Kumar, Prof. D.K. Chauhan, Prof. S.P. Mishra, Prof. Krishna Mishra, Prof. Azyz Sharafy (U.S.A.), Dr. B. Ray (U.S.A.), Dr. C.M. Nautiyal, Dr. A.R. Bahal, Dr. N.C. Mehrotra, Dr. A.K. Kukreja, Dr. Srubabati Goswami, Dr. Brajesh Dwivedi, Dr. O.P. Gupta, Dr. Pankaj Srivastava, Dr. Niraj Kumar, Dr. Manvendra Tripathi, Dr. Mukesh Khare, Dr. Ashish Tandon, Dr. Anjana Pandey, Dr. Mamta Srivastava, Dr. Ragini Mehrotra, Dr. V.C. Srivastava, Dr. Abhay Pandey, Dr. Md. Masood, Dr. K.P. Singh, Dr. Ravindra Dhar, Dr. Sharda Sundaram,Dr. Satyendra Singh, Dr. S.Kumar, Prof. S.D. Adhikari, Prof. B. Ramakrishna, Prof. K.P. Misra, Dr. Kamlesh Pandey, Dr. Deepak Chandra, Prof. R.K. Srivastava and Dr. Rajeev Srivastava.

IV. Teacher's Workshop

A. Workshop on Physics:

The state level Teacher's Workshop was held from February 24-26, 2008. More than 40 teachers from the Intermediates Colleges of U.P. participated in the workshop. On 24th February, the workshop was inaugurated by Prof. S.L. Srivastava ; Dr. O.P. Gupta, Retd. Teacher, demonstrated several experiments on laser beam effect on glass prism, mechanical physics, hydropressure etc. On next day, Dr. Brajesh Pandey from I.I.T. Kanpur, also demonstrated interesting experiments on center of mass and center of gravity, Pascal's law, functional theory, magnetic motor etc. After lunch, Prof. C.K. Dwivedi delivered a lecture on electrical efficiency and demonstrated the related experiments. On the same day, Dr. Md. Masood demonstrated water and food adulteration tests. On third day, Dr. Mrs. Srubabati Goswami delivered Dr. P. Sheel Memorial (Young Woman Scientist) Award Lecture (2006) on 'Dawn of a Nu-Era' and discussed about neutrino oscillations describing solar and reactor neutrinos. After lunch, Prof. H.C. Verma, from I.I.T. Kanpur, gave a talk on 'Challenges of the Present Education System' and discussed the role of teachers in education and moral building.

B. Workshop on Health:

On 27th February, 2008 a workshop on Health was organized. The purpose of this workshop, was to make the students and teachers aware about several diseases, so that they could adopt preventive and promotive health care strategies.

The following lectures and demonstrations were organized by medical practitioners and health experts- Dr. Mukesh Khare on Eye health, Dr. Ashish Tandon on Respiratory health, Dr. Manvendra Tripathi on Communicable diseases, Prof. U.C. Srivastava on Brain function, Dr. B.P. Agarwal on Cardiac health and Dr. Niraj Kumar on Nutrition. Cultural programmes as dance, drama and songs on health and environment were also presented by the students of Prestige Tutorial Inter College, Deoria, Tagore Public School and Umrao Singh Smarak Girls Inter College, Allahabad, which were enjoyed by all the participants.

REGULATIONS REGARDING THE NASI-SENIOR SCIENTIST PLATINUM JUBILEE FELLOWSHIP

The Academy has instituted the NASI Senior Scientist Platinum Jubilee Fellowship to utilize the services of the Fellows of the Academy who are active in high quality research in their specialized disciplines but have superannuated from their service. The Awardee shall be called NASI Senior Scientist.

Objectives - The objective is to utilize the expertise of NASI Fellows after superannuation primarily for research work in some R&D center/university/institute in India.

Eligibility & Age - Superannuated Fellows are eligible for consideration.

Number of Fellowships - The number of NASI Senior Scientists Platinum Jubilee Fellowship to be selected each year shall be decided by the Council from the panel recommended by the Committee, to be constituted by NASI. Usually, the number of Senior Scientists to be selected each year will be based on the availability of vacancies and funds available with the Academy. The maximum number at any given time shall not exceed five.

Tenure - The term of a NASI Senior Scientist position will be tenable initially for a period of three years, extendable for another two years after a review of the achievement of three year's works.

Emoluments* - The fellowship consists of Honorarium @ Rs.10,000/- per month and contingency grant @ Rs. 50,000/- per annum for meeting expenses on stationery, travel (within the country only). This also covers expenditure up to Rs. 10,000/- on communication needs. The honorarium is taxable, which will be deducted at source. One JRF (NET qualified) or SRF (with experience) will also be supported by resources from NASI.

Nominations - Nominations for the position shall be invited from the Fellows of the Academy in the first week of May preceding the year of the award.

The nomination papers duly completed in all respects, signed, and routed through the Head of the Institution, where a scientist intends to work, should be sent to the General Secretary, NASI, Allahabad so as to reach latest by 15th June.

Mode of Selection - The nominations received within the valid period shall be examined by a Committee constituted for the purpose. The recommendations of the Committee shall be considered and selection will be made by the Council at its meeting to be held in July.

Announcement - The names of NASI Senior Scientists selected shall be announced at the Annual General Body Meeting of the Academy.

Operation of Scheme - Funds for this scheme will be routed through the sponsoring institution.

Leave Rules - As per GOI norms.

Activities Report and Renewal of Scheme - Senior Scientist will submit an Annual Report of his/her research work in the prescribed format at the end of each Calendar year alongwith statement of expenditure for renewal and release of grant for the next year.

Final Technical Report - Senior Scientist is required to submit a consolidated technical report for the entire duration of the award at the end of tenure in the prescribed proforma.

Interaction with other Agencies - Senior Scientist is allowed to take up the sponsored Research and Consultancy within the norms of the host institute.

Obligations -

Host Institute shall submit the audited statement of accounts and utilization certificate from time to time to NASI. The Scheme will stand terminated from the date the Senior Scientist of NASI accepts any paid position elsewhere.

Acknowledgement of support in all publications as "Senior Scientist, NASI" should be made.

Final adjustment/settlement of accounts should be done within a period of 30 days of termination of Scheme.

All patent rights, designs and inventions derived from the research work of the NASI Senior Scientist shall be governed by the host institutional norms.

At the completion of the tenure of the NASI Senior Scientist, the awardee shall handover all permanent equipments purchased through the contingency funds to the institution where the work was undertaken.

They will be invited to submit paper/review to NASI journals.

These regulations may be revised or amended by the Council at any time.

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* to be enhanced and kept at par with INSA, New Delhi as and when implemented at INSA.

P.T.O

The National Academy of Sciences, India (NASI)
5, Lajpatrai Road, Allahabad – 211002

PROFORMA NOMINATION FORM

NASI – Senior Scientist Platinum Jubilee Fellowship

(10 copies of nomination and one set of reprints of 10 publications as given in para 8(d))

1. Name of the Nominee
2. Date & place of birth
3. (a) Designation and place of affiliation at the time of superannuation
   (b) Year of Superannuation
4. (a) Title of the proposed work
   (b) Name of the Institution where the Fellow intends to work **(The nomination paper must be routed through the Head of the Institution)**
5. Address with Tel / Fax / Email
   (a) Official, if any
   (b) Residence
   (any change may be immediately informed)
6. Academic qualifications (give in the form of a table from Bachelor's degree onwards)
7. Positions held (in chronological order)
8. (a) Summary of proposed work (give particulars in not more than 200 words);
   (b) Field of specialization
   (c) Area of research
   (d) List of publications in indexed journals (exclude abstract of presentations made in conferences/seminars, etc.; attach copies of not more than ten publications which you consider the best in the last 10 years);
   (e) List of patents granted/applied for, if any;
   (f) Details of technologies developed and commercialized/ ready for commercialization or have future potential;
   (g) List of books/ book chapters written;
   (h) Particulars of citation index of ten publications as submitted in 8(d).
9. Particulars of membership/fellowship in academies/societies/professional bodies.
10. Awards (give full particulars such as the agency/organization which gave the award, purpose and the nature of the award etc.)
11. Any other information in support of the nomination

Certificate

This is to certify that the above information about the nominee is correct.

(Signature of Nominator)
Name & Address

Date:
Place:

Endorsement by the Head of the Institution

Date:
Place:

Signature with Seal
Name & Address

List of those admitted as Members of NASI in the Year 2008

PHYSICAL SCIENCES

1 **Dr. Ranjit Kumar**
M.Sc.,Ph.D.
Lecturer in Chemistry,
Technical College,
Dayalbagh Educational Institute,
Agra - 282 005

2 **Dr. Ratikanta Mishra**
M.Sc.,Ph.D.
Scientific Officer, Chemistry Division,
Bhabha Atomic Research Centre,
Trombay,
Mumbai - 400 085

3 **Dr. P.A. Hassan**
M.Sc.,Ph.D.
Scentific Officer (F),Thermal and
Interfacial Chem. Sec.Chem. Division,
Bhabha Atomic Research Centre,
Trombay,
Mumbai - 400 085

4 **Dr. Man Mohan Srivastava**
M.Sc.,Ph.D.
Reader, Department of Chemistry,
Dayalbagh Educational Institute,
(Deemed University) Dayalbagh,
Agra - 282 005

5 **Dr. T.S. Basu Baul**
M.Sc.,Ph.D.
Senior Lecturer,
Department of Chemistry,
North-Eastern Hill University,
Umshing, NEHU Permanent Campus,
Shillong - 793 022

6 **Dr. S.B. Halligudi**
M.Sc., Ph.D.
Senior Scientist, Catalysis Division,
Natinal Chemical Laboratory,
Dr. Homi Bhabha Road,
Pune - 411 008

7 **Dr. Arvind Kumar Misra**
M.Sc.,Ph.D.
Reader, Department of Mathematics,
Banaras Hindu University,
Varanasi - 221 005

8 **Dr. K. Balachandran**
M.Sc.,B.Ed.,Ph.D.
Professor of Mathematics,
Department of Mathematics,
Bharthiar University,
Coimbatore - 641 046

9 **Dr. Somesh Kumar**
M.Sc, Ph.D.
Professor,
Department of Mathematics,
Indian Institute of Technology,
Kharagpur - 721 302

10 **Dr. D.K. Gupta**
M.Sc.,Ph.D.,
Professor, Department of Mathematics,
Indian Institute of Technology,
Kharagpur - 721 302

11 **Dr. P.K. Panigrahi**
Professor, Indian Institute of Science
Education and Research (IISER),
HC-VII, Sector III, Salt Lake,
Kolkata - -700 106

12 **Dr. S.P. Sharma**
M.Sc.(Tech.),Ph.D.
Associate Professor,
Department of Geology & Geophysics,
Indian Institute of Technology,
Kharagpur - 721 302

13 **Dr. Nibir Mandal**
M.Sc.,Ph.D.
Professor, Indian Institute of Science
Education and Research (IISER),
HC-VII, Sector III, Salt Lake,
Kolkata - 700106

14 **Prof. P.K. Ghosh**
M.E.,Ph.D.
Professor,
Deptt. of Metallurgical & Materials Engg,
Indian Institute of Technology Roorkee,
Roorkee - 247 667

15 **Dr. Deepankar Choudhury**
M.E., Ph.D.
Associate Professor,
Department of Civil Engineering,
Indian Institute of Technology Bombay,
Powai,
Mumbai - 400 076

16 **Prof. S.V. Kulkarni**
M.Tech., Ph.D.
Associate Professor,
Department of Electrical Engineering
Indian Institute of Technology Bombay,
Powai,
Mumbai - 400 076

17 **Prof. H.K. Pillai**
M.Tec.,Ph.D.
Associate Professor,
Deptt. of Electrical Engineering,
Indian Institute of Technology Bombay,
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